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INNEHÅLL.

DIE TERMINOLOGIE DER APOMIXIS-PROZESSE

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Bei vielen Pflanzen erfolgt Reproduktion ohne dass ein Sexualakt und ohne dass ein Kernphasenwechsel vorgekommen sind. Die Reproduktion kann da auf verschiedene Weise geschehen. Um hier Ordnung zu schaffen, sind besondere Termini eingeführt worden. Die Terminologie zeigt indessen eine immer grössere Neigung zu bedenklicher Verwirrung, weshalb mir eine kritische Prüfung derselben berechtigt erscheint. Die folgende Prüfung, Revision und Terminologie betreffen in erster Linie die höheren Pflanzen. Die Terminologie dürfte jedoch im grossen ganzen auch Geltung besitzen wie es sich um andere Organismen handelt.

Für sämtliche Fälle, wo ein Organismus sich fortpflanzt ohne dass Befruchtung erfolgt, ist der von DE BARY (1878) gegebene Ausdruck Apogamie angewandt worden. Fraglich ist indessen, ob DE BARY wirklich gemeint hat, dass der Ausdruck in diesem Sinne verwendet werden sollte. Aus seinen Definitionen und Beispielen geht das nicht klar hervor. Natürlich war die Problemstellung 1878 nicht dieselbe als heute, die Gesamtlage unseres Wissens bringt es mit sich, dass Definitionen und Exemplifikationen aus jenem Jahr heute oft nicht mehr anwendbar sind. Mir will es scheinen, als habe DE BARY mit seinem Ausdruck in erster Linie das Fehlen einer sexuellen Fortpflanzung bezeichnen wollen. DE BARYS Terminus ist später in verschiedener Weise verwendet worden: 1) zur Bezeichnung des Falles, wo Fortpflanzung mittels Samen geschieht ohne dass aber Befruchtung erfolgt ist 2) zur Bezeichnung der Bildung eines Sporophyten aus einem Gametophyten ohne dass Befruchtung erfolgt ist 3) zur Bezeichnung der Bildung eines Sporophyten aus einer somatischen Gametophytenzelle ohne dass Befruchtung erfolgt ist.

WINKLER (1908) führte den Ausdruck Apomixis ein um die geschlechtliche Fortpflanzung durch einen anderen nicht geschlechtlichen nicht mit Kern- oder Zellverschmelzung verbundenen Vermehrungsprozess zu bezeichnen. Der Ausdruck Apomixis ist noch später in genau so variabler Weise wie der Ausdruck Apogamie

verwendet worden. Ich benuge mich, hier ein Beispiel anzuführen.
DARLINGTON (1937, S. 434) schreibt: »Apomixis may be defined (follow-
ing WINKLER, 1908) as a system of reproduction having the external
character of sexual reproduction but omitting one or both of its essential
cellular processes. These are meiosis and fertilisation». Dies heisst doch
nicht WINKLER zu folgen.

Ist es erlaubt, auf diese Weise einmal definierte Ausdrucke ihre
Bedeutung ändern zu lassen? Ja, unter gewissen Umständen. Nam-
lich wenn die weitere Forschung Resultate ergibt — beispielsweise Ent-
deckungen, die mit sich bringen, dass in die Definition od r i. die erste
Definition und die ers e Exemplifikation ei hende Tatsachen ein-
ander widersprechen — ie ies verlangen, damit es iber up möglich
sein soll, mit der Term ie u aroeiten. Soweit al möglich muss
indessen auf die Priorität , T cksicht genommen werc en. Sinnände-
rungen ohne begrundeten Anlass sind unstatthaft Wa the Ände-
rung an der Bedeutung ine. Terminus vorgenon en o s eine
neue Definition geg werd n, wobei auch an ruc rt igt wer
den muss, dass die De nition neu ist. Was is gr gebu betrifft
so hat die weitere Forschung ka m in gns mas eine Ände-
rung der Bedeutung der Termin notwendig et cht rforderlich ist
vielmehr die Einfuhr g neuer ermint prs l en neb n den alten
anzuwenden sind.

Die gleichartige V nderung der Bedeutung der Termen Apogamie
und Apomixis beruht a zwei rch ingen l ein Termin
ist notwendig, um die igen alle u h umen wo Peproduktion
mittels Samen erfolgt, a r ohne dass Kerap enwechsl oder P uch-
tung geschieht (vgl. Da NGTONs Definition des Term as A mixis
siehe oben), 2) man hat ie Bildung res S rophyten aus einer fru-
heren Sporophytengeneration, eventuell ne eine Gametophytengene-
ration hin, mit der Bildung einer Sporophytengeneration aus einer
Gametophytengeneration vermengt n s amctophyter bildun aus
einem Sporophyten als Fortpflanzung des Sporophyten und Sporo
phytenbildung aus einem Gametophyten als Fortpflanzung des Gameto-
phyten bezeichnet. In diesem letzten hlichen Verfahren liegt zu
grossem Teil die Verwirrung, die in er er inologie eingetreten ist.
Man darf nicht den Ausdruck Fortpflanzung n dualistischer Bedeutung
verwenden. Unter Fortpflanzung duktion verstanden wer
den; die Bildung eines neuen Organismus aus einem gleichartigen, also
die Fortpflanzung des Sporophyten muss i der Bildung eines neuen

Sporophyten, und wenn Generationswechsel vorliegt, in einer solchen über einen Gametophytengeneration hin, bestehen.

1908 gab WINKLER eine Einteilung der verschiedenen Weisen, wie die apomiktische Vermehrung vor sich geht. Es scheint jedoch, als ob er dabei den Fehler begeht, auf den ich soeben hingewiesen habe. Er stellt nämlich drei Unterabteilungen, »vegetative Propagation, Apogamie und Parthenogenesis», einander gleich. Er definiert Apogamie: »die apomiktische Entstehung eines Sporophyten aus vegetativen Zellen des Gametophyten» und Parthenogenesis: »die apomiktische Entstehung eines Sporophyten aus einem Ei». Der erste der drei Ausdrücke bezieht sich auf eine Methode, nach welcher Fortpflanzung geschieht, die beiden anderen bezeichnen verschiedene Weisen, wie die Sporophytenbildung vom Gametophyten aus geschieht. WINKLER hat möglicherweise den Ausdruck Fortpflanzung in einer nachlässigen Weise benutzt. Wenn er hier von Fortpflanzung spricht, meint er wahrscheinlich nur Sporophytenbildung, sonst hätte er sich einer dualistischen Verwendung des Ausdruckes Apomixis schuldig gemacht. In dieser Weise — Apomixis = Sporophytenbildung ohne Befruchtung — hat offenbar EDMAN (1931) WINKLERs Arbeit aufgefasst. Die meisten Autoren sind aber dem Wortlaut der WINKLERschen Definition gefolgt. Es wäre jetzt sehr unpraktisch, anders zu tun. Folglich definiere ich: *Apomixis = Reproduktion ohne Befruchtung und also auch ohne Kernphasenwechsel.*

Es erhebt sich aber nun die Frage, welcher von den Termini zur Bezeichnung der Reproduktion ohne Befruchtung beizubehalten ist, Apogamie oder Apomixis. GUSTAFSSON (1935) antwortet: »As this term [scil. Apogamie] has in course of years been employed to denote different things, WINKLER's term is certainly to be preferred». Da jedoch der WINKLERsche Terminus ebensosehr wie der Terminus Apogamie »missbraucht» worden ist, verliert die von GUSTAFSSON angeführte Begründung ihren Wert. RENNER (1916) erklärt: »Nun sagt Gamie aber soviel wie Paarung, Mixis ist Mischung, Verschmelzung. Wir wissen, dass bei den höheren Pilzen allgemein der Geschlechtsvorgang zunächst in einer Kernpaarung besteht, auf die ganz spät erst die Kernverschmelzung folgt. Dieser zweite Schritt bleibt sogar ganz aus bei gewissen Uredineen — — —. Wir finden hier Gamie ohne Mixis, oder, um mit MAIRE zu sprechen, Apomixis». Schliesst man sich dieser letzteren Ansicht an, so müssten bei den höheren Pflanzen beide Ausdrücke, Apomixis und Apogamie, angewendet werden können, da ja weder Gamie noch Mixis vorliegt (man beachte jedoch Fälle von Pseudogamie!). RENNER lässt also die rein sprachliche Bedeutung der Termini

entscheidend sein. Dies ist jedoch kaum zweckmässig. Wenn ein Terminus einmal begrifflich definiert worden ist, muss er wenn möglich seine Bedeutung beibehalten, auch wenn sie, rein sprachlich gesehen, nicht völlig korrekt ist.

Da für den WINKLERschen Terminus eine genaue Definition vorliegt, da die Bedeutung des von DE BARY geschaffenen Terminus nicht völlig klar ist, und da der erstgenannte Terminus jetzt mehr in Gebrauch ist, ziehe ich es vor, den WINKLERschen Ausdruck zu benutzen, obwohl er jünger ist.

Individuen, bei denen die Reproduktion durch Apomixis bedingt ist, sind von TURESSON (1926) Apomikten genannt worden.

»Die drei Erscheinungen Apogamie, Aposporie und Nucellarembryonie können als verschiedene Fälle von apomiktischer Samenbildung (Embryobildung) zusammengefasst werden. Man könnte diese drei Fälle auch mit dem neuen Terminus *Agamospermie* bezeichnen», schreibt TÄCKHOLM (1922) in seiner wohlbekannten Abhandlung über *Rosa*. Wie TÄCKHOLM den Umfang der beiden Termini Apogamie und Aposporie abgrenzt, ist nicht deutlich erkennbar. Es scheint jedoch am ehesten, als wenn er dasselbe meinte wie ERNST (1918): mit Apogamie die Bildung eines Gametophyten aus einer Archesporzelle oder deren Derivaten und die Bildung eines Sporophyten aus diesem Gametophyten, ohne dass Kernphasenwechsel oder Befruchtung erfolgt ist, und mit Aposporie den analogen Fall, wo der Gametophyt aus einer rein somatischen Zelle hervorgegangen ist. *Unter Agamospermie wird* also, kurz gesagt, *verstanden Reproduktion mittels Samen, obwohl keine Befruchtung erfolgt ist.* Der Nutzen eines solchen Kollektivausdrucks liegt offen zutage. Auch solche Fälle, wo man nicht im Detail die Vorgänge im übrigen kennt, aber weiss, dass Reproduktion mittels Samen, obwohl ohne vorhergehende Befruchtung, geschieht, können auf diese Weise bezeichnet werden. Dass der Terminus wirklich nötig ist, zeigt die historische Entwicklung der Bedeutung der Termini Apogamie und Apomixis.

Zwei Typen von Apomixis können also aufgestellt werden: Agamospermie und Reproduktion ohne Samen.

Individuen, die sich durch Agamospermie reproduzieren, können Agamospermen genannt werden.

Die agamospermische Entwicklung tritt in ihrer einfachsten Form hervor, wenn Nucellarembryonie (embryonia estrasaccale; CHIARUGI) vorliegt. Hier entsteht direkt aus einer Sporophytenzelle, näher bestimmt aus einer Nucellus- oder Integumentzelle (oder Plazentazelle?),

ein Embryo, ein neuer Sporophyt. Nucellarembryonie ist nicht mit
Generationswechsel verbunden. Alle anderen Fälle von Agamospermie
sind durch Generationswechsel — nicht aber Kernphasenwechsel —
gekennzeichnet. Ein Terminus für diese Typen von Agamospermie
fehlt. Das Bedürfnis nach einem solchen liegt jedoch vor. *Agamogonie*
scheint mir geeignet. Zwar sind hier schon früher Ausdrücke zur An-
wendung gekommen, nämlich Parthenogenesis, Apogamie und Apo-
sporie. Wo dies geschehen ist, sind dieselben aber in fehlerhafter Weise
verwendet worden (siehe unten).

Individuen, die sich durch Agamogonie reproduzieren, können als
Agamogonen bezeichnet werden.

Bei agamogonischer Entwicklung spielen sich zwei verschiedene
Prozesse ab: Gametophytenbildung, die nicht mit Chromosomenzahl-
reduktion verbunden ist, und Sporophytenbildung, die nicht mit Be-
fruchtung verbunden ist. Jeder dieser beiden Prozesse kann auf ver-
schiedene Weise vor sich gehen. Um hier Ordnung zu schaffen, ist es
notwendig, bestimmte Termini zu gebrauchen. Solche sind auch vor-
handen. Eine scharfe Angabe ihrer Bedeutung ist jedoch notwendig,
da sie in verschiedenem Sinne angewandt worden sind.

Für die zwei verschiedenen Weisen, auf welche Sporophyten-
bildung ohne Befruchtung erfolgen kann, benutzte WINKLER die Ter-
mini Parthenogenesis und Apogamie. Seine Definitionen sind oben
referiert. Sein Terminus Apogamie ist aber von RENNER (1916) durch
den neuen Ausdruck Apogametie ersetzt worden, den eine Reihe von
Forschern akzeptiert haben (z. B. ROSENBERG, 1930; EDMAN, 1931),
deshalb nämlich, weil der Ausdruck Apogamie in so vielen verschiede-
nen Bedeutungen gebraucht worden ist. Wenn die Zelle, die den Sporo-
phyten liefert, die unreduzierte Chromosomenzahl besitzt, spricht man
von somatischer, diploider (Diplo-) oder zygoider, wenn sie die redu-
zierte Zahl hat, von generativer, haploider (Haplo-) oder azygoider
Parthenogenesis bzw. Apogametie.

Die späteren Prozesse bei agamogonischer Entwicklung erfolgen
also durch (Diplo-) *Parthenogenesis = Sporophytenbildung ohne Be-
fruchtung aus einem* (unreduzierten) *Ei oder durch* (Diplo-) *Apoga-
metie = Sporophytenbildung ohne Befruchtung aus einer* (unreduzierten)
vegetativen Gametophytenzelle.

Für die verschiedenen Weisen, auf welche die unreduzierten Ga-
metophyten gebildet werden können, führte WINKLER keine Termini
ein. Er schreibt nur (1908, S. 71): ›Wir bezeichnen als somatische
Parthenogenesis die ohne vorhergehende Befruchtung erfolgende Ent-

wicklung einer Eizelle zum Embryo, deren Kern von vornherein die diploide Chromosomenzahl führt. Da nun die Eizelle des Gametophyten normalerweise einen haploidchromosomigen Kern besitzt, so ist es klar, dass auch die somatische Parthenogenesis wie die somatische Apogamie [= 'Apogametie'] *mit einem Vorgang verbunden sein muss* [1], der den Kernen der Gametophytenzellen anstatt der für sie typischen haploiden die diploide Chromosomenzahl verleiht. Es sind zwei Modalitäten denkbar, durch die das erreicht werden kann — — —. Erstens *kann sich die somatische Parthenogenesis mit Aposporie kombinieren* [1], d. h. also, es kann eine normale, also diploidchromosomige Sporophytenzelle unmittelbar zum Gametophyten auswachsen. *Zweitens aber kann der Gametophyt auf dem gewöhnlichen Wege, also aus einer Spore hervorgehen, wobei aber die sonst bei der Sporenbildung stattfindende Reduktionsteilung unterbleibt*» [1].

WINKLER versteht also unter Parthenogenesis und Apogamie [= Apogametie] bei agamogonischer Entwicklung nur die spätere Phase. Das geht deutlich aus dem obigen Zitat und aus seiner ganzen Arbeit hervor. GUSTAFSSON (1935) schreibt jedoch: »In this work parthenogenesis is thus defined, in accordance with WINKLER and ROSENBERG, as the process by means of which an azygoid (haploid) or a zygoid (diploid) egg-cell is produced and then develops without any fusion of nuclei and cells». In den Begriff Eizellbildung schliesst GUSTAFSSON hier auch die Bildung des Gametophyten aus dem Sporophyten ein. Parthenogenesis wäre demnach = eine Form der ganzen agamogonischen Entwicklung. Dass er das auch meint, geht aus den Darlegungen in seiner Abhandlung hervor. GUSTAFSSON lässt also den Terminus Parthenogenesis das bezeichnen, wofür ERNST (1918) praktisch den Ausdruck ovogene Apogamie verwendete (siehe oben). GUSTAFSSON hat offenbar an WINKLERs Gleichstellung von Sporophytenreproduktion mit Bildung des Sporophyten aus einem Gametophyten Anstoss genommen. Er hat WINKLER eine andere Definition in den Mund gelegt. Sein Verfahren stellt zwar eine Lösung dar, es bringt aber gleichzeitig Kollisionen mit anderen Begriffen mit sich und ist in der praktischen Anwendung sehr unbequem. Die Erscheinung, die sich bei *Ochna multiflora* (FRANCINI, 1928) findet, Bildung unreduzierter Embryosäcke, aber nicht Bildung von Embryonen aus diesen, würde bei dieser Terminologie als abgebrochene Parthenogenesis zu bezeichnen sein. Nahezu unmöglich ist es, die Terminologie anzuwenden, wenn es sich um haploide Parthenogenesis handelt.

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[1] Von mir kursiviert.

Dieselbe Schwierigkeit wie GUSTAFSSON hatte EDMAN (1931) früher. Er wählte den entgegengesetzten Weg, um über sie hinwegzukommen. Er wendet die Termini Parthenogenesis und Apogametie in ihrer eingeschränkten Bedeutung an, den Terminus Apomixis aber als eine Kollektivbezeichnung für die beiden vorgenannten Vorgänge (sehr logisch!), also für Sporophytenbildung, nicht für Reproduktion ohne Befruchtung.

Bei der agamogonischen *Antennaria alpina* bezeichnet JUEL (1900) die Embryosackbildung als homolog mit Aposporie. Der Terminus ist früher vor allem inbezug auf Farne angewandt worden, wo Gametophyten direkt aus dem Soma des Sporophyten, ohne Reduktionsteilung, gebildet werden. WINKLER (1908) will offenbar den Terminus Aposporie inbezug auf die Angiospermen für den damals nur bei *Hieracium* von ROSENBERG (1907) angetroffenen Fall reservieren, wo ein Gametophyt sich direkt aus einer rein somatischen Zelle in der Samenanlage, also nicht aus einer E.M.Z. entwickelt. CHIARUGI (1926) verwendet den Ausdruck sowohl für den Fall, wo ein unreduzierter Embryosack aus einer Archesporzelle gebildet wird (»Aposporia goneale»), als auch für den Fall der Entstehung aus einer rein somatischen Zelle (»Aposporia somatica»), ein Verfahren, das ROSENBERGs (1930) Billigung findet. Wie dem nun auch sei, so muss doch der Terminus Aposporie für die frühere Phase der agamogonischen Entwicklung, für die Bildung des Gametophyten, reserviert bleiben. Hiergegen haben ERNST (1918) und GUSTAFSSON (1935) verstossen. Dass ERNSTs und GUSTAFSSONs Terminologien sehr unpraktisch und daher zu verwerfen sind, geht aus dem Resultat hervor, zu dem ERNST gelangt, wenn er wirklich die Begriffe zu definieren versucht (vgl. EDMAN, 1929). Man vergleiche ERNSTs Tabelle (1918, neben S. 596); dort steht als Unterrubrik zu »Fortpflanzung und Vermehrung unter Ausschaltung des Befruchtungsprozesses»: »*Apogamie* = Bildung von Gametophyten und Keimen nach somatisch durchgeführter Teilung der Sporen-(Embryosack-)mutterzellen. *Apospore* = Bildung von Gametophyten unter Umgehung der Sporenbildung. Fortpflanzung der Gametophyten durch ovogene oder somatische *Apogamie*». Das einzig Vernünftige ist, den Terminus Aposporie nur eine gewisse Form der Gametophytenbildung, bei Agamogonie nur der früheren Phase in der Entwicklung bezeichnen zu lassen. Verfährt man wie GUSTAFSSON und ERNST und geht man dann unbekümmert seinen Weg weiter, so gelangt man zu Bezeichnungen und Sätzen wie »parthenogenetische Meiosis», »parthenogenetische Prophase» und als Gegensatz zu diesen »sexuelle Meiosis», »sexuelle Prophase» und ähnliche

Ausdrücke, die in GUSTAFSSONs Arbeiten (vgl. z. B. GUSTAFSSON, 1939 a)
sehr häufig vorkommen.

Für alle die Fälle, wo ein Gametophyt ohne Reduktion der Chro-
mosomenzahl gebildet wird, führte RENNER (1916) den Terminus Apo-
meiosis ein.

Bei agamogonischer Entwicklung ist also Apomeiosis die erste
Phase. Einen Fall von Apomeiosis stellt Aposporie dar, wie ist nun aber
der letzte Terminus abzugrenzen? Es ist im vorstehenden gezeigt
worden, dass die Meinungen in dieser Beziehung auseinandergehen.
Alle, die unter Aposporie nur eine Form von Gametophytenbildung ver-
stehen, sind sich jedoch darüber einig, dass der Terminus für die Fälle
benutzt werden muss, wo man nicht von Sporen sprechen kann, d. h.
wo der Embryosack und dessen Mutterzelle nur rein somatische Tei-
lungen erfährt. Die Entscheidung liegt also bei der Frage, wann eine
Teilung als somatisch aufzufassen ist und wann nicht. Wir wissen
nun, dass Zwischenformen sich finden. HOLMGREN (1919) wies darauf
hin, dass bei agamogonischen *Erigeron*-Arten bei der Embryosackbil-
dung eine Äquationsteilung durchgeführt wird, dass aber die Chromo-
somen dann die kontrahierte Gestalt haben, die die Meiosis kennzeichnet
— die pseudohomotypische Teilung (GUSTAFSSON), der *Erigeron*-Typ
(BERGMAN). Diese Teilung, zu deren Kenntnis GUSTAFSSON fleissig bei-
getragen hat, muss meines Erachtens als eine Zwischenform zwischen
Meiosis und Mitose betrachtet werden. Wenigstens während der Pollen-
bildung werden bei *Wikstroemia* (FAGERLIND, 1940) Zwischenformen
beobachtet zwischen diesem Teilungstyp und einem Teilungstyp, der
jedenfalls dem Äussern nach identisch mit einer Mitose ist — vgl. auch
verschiedene *Hieracium*-Arten (ROSENBERG, 1927; GENTSCHEFF, 1937).
Als solche stark (vollständig?) nach dem mitotischen Stadium hin ver-
schobene Zwischentypen sind sicher die E.M.Z.-Teilungen aufzufassen,
die von GUSTAFSSON als *Hieracium*-Typ und von BERGMAN als *Eupato-
rium*-Typ bezeichnet worden sind. Eine Reihe Zwischenformen finden
sich wahrscheinlich auch zwischen der pseudohomotypischen Teilung
und der stark asyndetischen Meiosis, die wenigstens während der Pollen-
bildung bei vielen Agamogonen gewöhnlich ist (vgl. z. B. *Wikstroemia*;
FAGERLIND, 1940). Schon diese Asyndese ist meines Erachtens als ein
Schritt auf dem Wege zur Somatisierung der Teilung aufzufassen (vgl.
FAGERLIND, 1940). Die Teilung, die zur Bildung eines Embryosacks
bei den Agamogonen führt, kann demnach verschiedene Lagen auf einer
von 0 % bis zu 100 % laufenden Skala einnehmen, auf der der 0-Punkt
eine reine Meiosis und der 100-Punkt eine reine Mitose bezeichnet.

Wenn die Teilung rein somatisch ist, kann man nicht von dem Vorkommen von Sporen sprechen. Hier ist der Terminus Aposporie zur Bezeichnung der Gametophytenbildung berechtigt. Dieser Fall ist verwirklicht, wenn der Gametophyt aus einer rein somatischen Zelle hervorgegangen ist — Aposporia somatica (CHIARUGI, 1926) — und wenn die Archesporzelle eine rein somatische Teilung durchgemacht hat. Für den letzteren Fall ist CHIARUGIS Terminus Aposporia goneale zu reservieren. Die beiden Ausdrücke sind von ROSENBERG (1930) mit somatische bzw. generative Aposporie übersetzt worden. Die somatische und die generative Aposporie gehen ohne Grenze ineinander in den Fällen über, wo man nicht entscheiden kann, ob eine Zelle als Archesporzelle oder als somatische Zelle aufzufassen ist (besonders ist dies schwierig in den peripheren Teilen eines mehrzelligen Archespors). Bei gewissen Arten der Gattungen *Elatostema* und *Pellionia* (eigene unveröffentlichte Studien) lässt sich ebenfalls eine distinkte Grenze nicht ziehen. Hier ist nämlich eine Archesporzelle oft überhaupt nicht ausdifferenziert. Eine einzige somatische Zelle im Nucellus wächst zu einem Embryosack aus, sie kann ja als homolog mit einer Archesporzelle betrachtet werden. Bei anderen Arten ist die Archesporzelle mehr oder weniger differenziert, sie entwickelt sich zu einem reduzierten oder unreduzierten Embryosack, oder sie degeneriert, wobei Embryosäcke von rein somatischen Zellen gebildet werden.

EDMAN (1931) vermutet, dass, wenn ein unreduzierter Embryosack aus einer E.M.Z. oder aus einer Dyadenzelle gebildet wird, die Gametophyteninitiale stets als eine unreduzierte Spore aufzufassen sei. Wenn Restitutionskerne bei der Pollenbildung die Bildung von unreduzierten Pollen verursacht haben, spricht man fortgesetzt von Sporen. Dann müsse man auch von Sporen sprechen, wenn unreduzierte Derivate durch die Teilung des E.M.Z.-Kerns erhalten werden, meint er. Für den Fall: Gametophytenbildung aus einer E.M.Z. durch eine Teilung, die nicht Reduktion der Chromosomenzahl bewirkt, führt EDMAN den Terminus Diplosporie ein. Er bemerkt zu diesem Ausdruck, dass er eine Verbindung der Wörter »diploide Spore» darstellt. Der Terminus ist gut. Er ist aber — meine ich — nur für den Fall zu gebrauchen, wo die Teilung der E.M.Z. eine wirkliche Meiosis ist und die Chromosomenzahl also durch Restitutionskernbildung bewahrt ist. Ich definiere also Diplosporie etwas anders als EDMAN (siehe unten).

Die Frage ist nun die: Gibt es überhaupt — unter Berücksichtigung der oben aufgestellten Forderungen — generative Aposporie und Diplosporie? Aus den obigen Darlegungen ist es klar, dass diese Ter-

TABELLE 1. Verschiedene Arten von Reproduktion bei den höheren Pflanzen.

Befruchtung und Kernphasenwechsel gehen in die Reproduktionsprozesse ein

Reduktion der Chromosomenzahl leitet die Gametophytenbildung ein

Reduzierter Gametophyt — Sporophyt

Die Sporophytenbildung erfolgt ohne Befruchtung

Die Sporophytenbildung wird durch Befruchtung eingeleitet

(Haplo-)Apogametie. Die Initialzelle ist nicht eine Gametenzelle	(Haplo-)Parthenogenesis. Die Initialzelle ist eine Gametenzelle

Apomixis (WINKLER, 1908)
Reproduktion ohne Befruchtung und Kernphasenwechsel

Chromosomenzahlreduktion kommt bei der Gametophytenbildung nicht vor. — Apomeiosis (RENNER, 1916)

Aposporie Die erste Teilung der Mutterzelle ist stark mitotischen Charakters	Semiaposporie Die erste Teilung der Mutterzelle ist pseudohomotypisch	Diplosporie Die erste Teilung der Mutterzelle ist stark meiotischen Charakters
Somatische Aposporie Die Mutterzelle ist eine somatische Zelle	Generative Aposporie Die Mutterzelle ist eine Archesporzelle	

Agamospermie (TACKHOLM, 1922)
Reproduktion durch Samen

Unreduzierter Gametophyt — Sporophyt

Agamogonie Generationswechsel liegt vor

Die Sporophytenbildung erfolgt ohne Befruchtung

Die Sporophytenbildung wird durch Befruchtung eingeleitet

(Diplo-)Apogametie Die Initialzelle ist nicht eine Gametenzelle	(Diplo-)Parthenogenesis Die Initialzelle ist eine Gametenzelle

Generationswechsel liegt nicht vor

»Nucellarembryonie«, Generationswechsel verschiedene Arten von vegetativer Reproduktion)

Die Reproduktion erfolgt nicht mittels Samen (viele Arten von vegetativer Reproduktion)

mini für zwei extreme Erscheinungen angewandt werden müssen, die durch eine ganze Kette von intermediären Typen verbunden sind. Theoretisch ist es also unmöglich, eine Einteilung vorzunehmen. Ich schlage daher einen Kompromiss dem Nachstehenden gemäss vor:

Aposporie = Bildung eines unreduzierten Gametophyten aus einer Zelle, die eine relativ stark mitotisch betonte Teilung erfährt.

Diplosporie = Bildung eines unreduzierten Gametophyten aus einer Archesporzelle, die eine Teilung von relativ stark meiotischem Charakter erfährt, welche zur Bildung eines Restitutionskerns führt.

Zwischen diesen beiden Typen gibt es nun intermediäre Fälle. Ein markanter Fall dieser Art ist es, wenn die Teilung der E.M.Z. pseudohomotypisch gewesen ist. Für diesen Fall schlage ich den Terminus *Semiaposporie* vor. Die drei Fälle Aposporie, Semiaposporie und Diplosporie gehen ohne Grenze ineinander über, wie es auch die drei Teilungstypen Mitose, pseudohomotypische Teilung und Meiosis tun.

Hiermit ist der erste Teil der terminologischen Diskussion abgeschlossen. Das Resultat ist eine Terminologie, die meines Erachtens den Anforderungen an Logik genügt, und mit der sich praktisch arbeiten lässt. Sie wird durch Tabelle 1 veranschaulicht. Die gestrichelten Grenzen geben an, dass die Abgrenzung diffus ist, dass Zwischentypen möglicherweise vorhanden sind. Die Verlängerung gewisser Kolumnen über die eigentliche Tabelle hinaus gibt Fälle an, die weder als Agamospermie noch als das Gegenteil davon — Gamospermie — bezeichnet werden können. Diese Zwischenformen betreffen die Fälle, in denen ein unreduzierter Embryosack befruchtet wird, oder in denen ein reduzierter Embryosack einen Sporophyten ohne Befruchtung entwickelt. Termini, die den Entwicklungszyklus hier bezeichnen, sind kaum vonnöten.

In der Tabelle und im Text sind auch für die Agamogonen die Termini Sporophyt und Gametophyt verwendet worden. Möglicherweise kann, da Sporen hier nicht immer gebildet werden, die Richtigkeit des Verfahrens diskutabel erscheinen.

Diejenigen Fälle bei den Agamogonen, wo der Gametophyt aus einer Archesporzelle hervorgeht, sind früher in folgender Weise eingeteilt worden: *Antennaria*-Typ — die E.M.Z. entwickelt sich direkt zum Embryosack, *Taraxacum*-Typ — die E.M.Z. wird zu einer Zellendyade, der Gametophyt entsteht aus einer der Tochterzellen, *Alchemilla*-Typ — die E.M.Z. wird zu einer Zellentetrade, der Gametophyt entsteht aus einer der Tochterzellen (vgl. ROSENBERG, 1930). Der *Alchemilla*-Typ ist indessen auf Grund falscher Schlüsse aufgestellt worden, er ist

also zu streichen (vgl. ROSENBERG, 1930; GUSTAFSSON, 1935; LILJEFORS, 1934 und dort angeführte Literatur).

Dass die Verschiedenheit der Chromosomenform bei den Äquationsteilungen, die während der semiaposporischen und der aposporischen Entwicklung stattfinden, als Einteilungsgrund verwendet werden muss, wird von BERGMAN wie auch von GUSTAFSSON betont. BERGMAN nennt den letzteren Entwicklungstyp *Eupatorium*-Typ, GUSTAFSSON *Hieracium*-Typ. Beide Autoren erklären, dass eine Einteilung, die davon ausgeht, ob die E.M.Z. zwei Tochterzellen gebildet hat oder aber ungeteilt geblieben ist, weniger berechtigt ist als eine Einteilung nach dem Kernteilungstyp. Dieser Ansicht muss man beistimmen. Es handelt sich ja um etwas so Wesentliches wie die Teilung selbst, die zur Gametophytenbildung führt. Die andere Einteilung gründet sich auf das Resultat der Teilung. Der erstere Einteilungstyp ist daher bei dem Schema in Tabelle 1 zur Verwendung gekommen. Was ich dort generative Aposporie nenne, ist also identisch mit BERGMANs *Eupatorium*- und GUSTAFSSONs *Hieracium*-Typ.

Es fragt sich nun: Stehen Diplosporie, Semiaposporie und generative Aposporie irgendwie in Zusammenhang mit *Taraxacum*-Typ und *Antennaria*-Typ? Bei den Gamospermen gibt es drei verschiedene Typen hinsichtlich der Derivate des E.M.Z.-Kerns: den Normaltyp — die vier Sporenkerne sind durch Wände geschieden; den bisporischen Typ — nur eine Wand ist vorhanden, 2 Sporenkerne sind im Embryosack enthalten; den tetrasporischen Typ — Wände werden überhaupt nicht gebildet, 4 Sporenkerne sind im Embryosack enthalten (über diese Typen siehe des näheren FAGERLIND, 1937—1939 c, und dort angeführte Literatur). Besteht ein Zusammenhang zwischen diesen Typen und dem *Taraxacum*- bzw. *Antennaria*-Typ?

GUSTAFSSON (1935) versuchte die beiden Fragen zu beantworten. Sein Resultat war: »By means of this classification it has been shown that the division into *Antennaria*, *Taraxacum* and *Alchemilla* schemes does not correspond to or is even analogous with the division into *Lilium*, *Scilla* and Normal types for sexual plants, that the wall formation on which this division is based does not correspond to the method of division in the E.M.C. — — —». Es besteht indessen sicher ein gewisser Zusammenhang, was sich klar ergibt, wenn man die Verhältnisse näher betrachtet, die in der nachstehenden Tabelle 2 uns entgegentreten.

In die Tabelle sind nur Fälle aufgenommen worden, die als ziemlich sicher angesehen werden können. Auch die unsicheren Fälle in die Diskussion einzubeziehen, wäre zwecklos. Die Angaben (sie stehen

im grossen ganzen in Übereinstimmung mit GUSTAFSSONs Zusammen-
stellung) sind teils dem Text der Originalarbeiten, teils den Illustrationen
derselben entnommen, teils sind sie Resultate eigener unveröffentlichter
Studien (*Elatostema* und *Pellionia*).

GUSTAFSSON wies darauf hin, dass, wenn die Teilung stark somati-
schen Charakter gehabt hat, stets *Antennaria*-Typ die Folge ist. Dies er-
gibt sich auch aus der Sektion I in meiner Tabelle, wenn man von den
Variationen absieht. Aus der Sektion II geht hervor, dass, wenn Resti-
tutionskerne oder pseudohomotypische Teilung zur Entstehung des
unreduzierten Gametophyten geführt haben, zumeist *Taraxacum*-Typ
die Folge ist. Dagegen, dass dies eine generelle Regel ist, spricht die
Sektion III der Tabelle.

Diese dritte Sektion enthält Fälle, wo der Teilungstyp derselbe wie
in Sektion II, das Resultat aber dasselbe wie in Sektion I gewesen ist.
Die Erklärung liegt sehr nahe. Unter diesen Fällen befindet sich
Erigeron Karwinskianus v. *mucronatus*. Bei dieser Art liegt Variation
vor, reduzierte Embryosäcke können gebildet werden (CARANO, 1919,
1921; vgl. FAGERLIND, 1939 b). Diese Embryosäcke sind von tetraspori-
schem Typ. Wenn eine solche Pflanze unreduzierte Embryosäcke aus-
bildet, muss das Resultat unabhängig von dem Charakter der Teilung
Antennaria-Typ sein. Innerhalb der Gattung *Erigeron* sind Arten, die
dem bi- und tetrasporischen Typ folgen, gewöhnlich. Dass *E. annuus*
und *ramosus* dem *Antennaria*-Typ folgen, obwohl ihre Teilung nicht
als stärker somatisiert bezeichnet werden kann, kann da nicht weiter
erstaunlich erscheinen. TAHARA (1921) zeigt, dass bei *E. annuus* tem-
poräre Zellplattenbildung zwischen den Tochterkernen stattfindet. Es
ist dies ein Zug, der oft für tetrasporische Embryosäcke charakteristisch
ist. (Temporäre Zellplatten können jedoch auch während Teilungen
im Embryosack vorhanden sein!) *Ixeris dentata* verhält sich wohl auf
dieselbe Weise, die temporäre Membranbildung ist auch hier beobachtet
worden. Die Gametophytogenese normaler *Ixeris*-Arten ist nicht be-
kannt. Die nächsten Verwandten, die *Lactuca*-Arten, zeigen Normaltyp.
Da das Vorliegen bald von Normal-, bald von tetrasporischem oder bi-
sporischem Typ bei einander sehr nahestehenden Arten nicht unge-
wöhnlich ist (vgl. FAGERLIND, 1938), braucht der *Ixeris*-Fall nicht als
gegen meine Schlussfolgerung sprechend angesehen zu werden.

Die Fälle in der dritten Sektion der Tabelle würden also dem
Antennaria-Schema folgen, obwohl die erste Teilung der E.M.Z. nicht
somatisch gewesen ist, da ihre Entwicklung auf eine Weise geschehen
ist analog derjenigen der bi- oder tetrasporischen und nicht der mono-

TABELLE 2

Fall	Embryosacktyp bei nachstverwandten, bekannten amphimiktischen Arten	Teilung I der E. M. Z.				Resultat			Autoren
		± abnorme Meiosis	Restitutionskernbildung	Pseudohomotypische Teilung	Somatische Teilung	± abnorme »Tetraden«	Taraxacum-Typ	Antennaria-Typ	
Antennaria spp...	N	+			+	zuwei-len		+	BERGMAN, 1936 c; JUEL, 1900; STEBBINS, 1932.
Arnica alpina ...	N				+			+	AFZELIUS, 1936.
Elatostema und Pellionia spp...	N				+			+	STRASBURGER, 1910. Eigene unveröffentlichte Studien.
I Eupatorium glandulosum ...	N				+			+	HOLMGREN, 1919.
Hieracium spp...	N	?	?	zuwei-len	+	zuwei-len	zuwei-len	+	BERGMAN, 1935 b; GENTSCHEFF, 1937; GUSTAFSSON, 1935; ROSENBERG, 1917, 1927.
Poa spp. ...	N	selten			+	selten		+	KIELLANDER, 1937; nebst von ihm mitgeteilten, nicht veröffentlichten Resultaten.

Gruppe	Art	Typ							Literatur
	Balanophora japonica	N?		+			+	+	KUWADA, 1928.
II	*Chondrilla* spp.	N	selten?	+	+	selten?	+	+	PODDUBNAJA-ARNOLDI, 1933; ROSENBERG, 1912; (vgl. GUSTAFSSON, 1935).
	Taraxacum spp.	N		+	—		ca. 50% ca. 50%	+	GUSTAFSSON, 1935; OSAWA, 1913.
	Wikstroemia viridiflora	N	selten?	+	zuweilen	selten?	?	+	FAGERLIND, 1939d; STRASBURGER, 1909. (WINKLER, 1906).
III	*Erigeron annuus* und *ramosus* (vgl. SÖDERBERG, 1929)	N 2-sporisch 4-sporisch	+		selten		+	+	GUSTAFSSON, 1935; HOLMGREN, 1919; McDONALD, 1927; TAHARA, 1921.
	Erigeron Karwinskianus v. *mucronatus*	4-sporisch	+	+			+	+	CARANO, 1921.
	Ixeris dentata	?	+	+			+	+	OKABE, 1932.

sporischen Schemata. Bei den Amphimikten erfolgt in der Regel (nicht immer!) Vakuolenbildung und kräftiges Wachstum des Gametophyten erst, nachdem vier Sporenkerne gebildet worden sind, gleichgültig welcher Entwicklungsmodus vorgelegen hat (vgl. FAGERLIND, 1937—1939). In den Fällen, die in der ersten Sektion der Tabelle 2 aufgeführt sind — Fälle mit generativer Aposporie — geschieht Vakuolenbildung schon in der E.M.Z. vor der somatischen Teilung, in den Fällen der zweiten Sektion — Fälle mit Semiaposporie — erst in der Dyadenzelle, also nach der ersten Teilung, aber vor der ersten somatischen Teilung. In diesen sämtlichen Fällen konnte die Entwicklung als analog dem Normaltyp verlaufend angesehen werden. Besässen dieselben Regeln Geltung, wenn die Entwicklung analog den bi- und den tetrasporischen Schemata verläuft, so wäre der Zeitpunkt der Bildung der Vakuolenbildung von entscheidender Bedeutung für die Beurteilung der drei Fälle in der dritten Sektion der Tabelle 2. Die drei Sektionen in Tabelle 3 zeigen, wie die Entwicklungsphasen aufeinander folgen müssen, wenn »Sporie», Diplosporie, Semiaposporie und Aposporie vorliegen und die Entwicklung analog den monosporischen (Sektion I), den bisporischen (Sektion II) und den tetrasporischen Schemata (Sektion III) verläuft.

Wenn die drei Fälle in der dritten Sektion der Tabelle 2 so zu erklären sind, wie ich oben geltend gemacht habe, müssen sie Fälle vom *Antennaria*-Typ repräsentieren, wo die Vakuolenbildung erst eintritt, wenn zwei Kerne in der »E.M.Z.» vorhanden sind. Sowohl TAHARA als auch HOLMGREN sagen es im Text und zeigen es durch Abbildungen, dass die Phase der grossen Volumzunahme und die Vakuolenbildung bei *Erigeron annuus* bzw. *ramosus* erst eintreten, nachdem zwei Kerne gebildet worden sind. Für *Ixeris* geht dasselbe aus OKABES Illustrationen hervor. CARANO gibt einige Bilder von *Erigeron Karwinskianus,* die ebenfalls in guter Übereinstimmung mit dem zu Erwartenden stehen. Die komplizierten Verhältnisse hier machen indessen den Fall ein wenig unsicher, aber eben hier liegt ja ein vollgültiger Beweis dafür vor, dass die Entwicklung analog den tetrasporischen Schemata erfolgt. Die Richtigkeit meiner Ansicht von dem Zusammenhang zwischen *Taraxacum*-Typ und *Antennaria*-Typ einerseits und Diplosporie und Semiaposporie bzw. Aposporie andererseits ist somit einwandfrei erwiesen.

Aus Tabelle 2 ist ersichtlich, dass eine ganze Reihe Variationen bei ein und demselben Fall vorkommen. In mehreren der Variationen tritt dieselbe Übereinstimmung zutage, auf die ich soeben hingewiesen habe. Es lässt sich demnach der Schluss ziehen: *In den meisten Fällen ist das*

TABELLE 3.

Wachstum und Vakuolenbildung — Erste somatische Teilung u. s. w.

	Heterotypische Teilung	Zellen-Dyade Wandbildung	Homotypische Teilung	Zellen-Tetrade Wandbildung
Monosporische Typen	Heterotypische Teilung	Zellen-Dyade Wandbildung	Homotypische Teilung	Zellen-Tetrade Wandbildung
I — Diplosporie (*Taraxacum*-Typ)	Semiheterotypische Teilung und Restitutionsbildung	1-kernige Zelle	Homotypische Teilung	Zellen-Dyade Wandbildung
Semiaposporie (*Taraxacum*-Typ)	———	→	Pseudohomotypische Teilung	Zellen-Dyade Wandbildung
Aposporie (*Antennaria*-Typ)	———			↑
Bisporische Typen	Heterotypische Teilung	Zellen-Dyade Wandbildung	Homotypische Teilung	Zellen-Dyade mit 2-kern. Zelle(n)
II — Diplosporie (*Antennaria*-Typ mit »später« Vakuolenbildung)	Semiheterotypische Teilung und Restitutionskernbildung	1-kernige Zelle	Homotypische Teilung	2-kernige Zelle
Semiaposporie (*Antennaria*-Typ mit »später« Vakuolenbildung)	———		Pseudohomotypische Teilung	2-kernige Zelle
Aposporie (*Antennaria*-Typ)	———	→		↑
Tetrasporische Typen	Heterotypische Teilung	2-kernige Zelle	Homotypische Teilung	4-kernige Zelle
III — Diplosporie (*Antennaria*-Typ mit »später« Vakuolenbildung)	Semiheterotypische Teilung und Restitutionskernbildung	1-kernige Zelle	Homotypische Teilung	2-kernige Zelle
Semiaposporie (*Antennaria*-Typ mit »später« Vakuolenbildung)	———		Pseudohomotypische Teilung	2 kernige Zelle
Aposporie (*Antennaria*-Typ)	———	→		↑

*Vorliegen des Antennaria-Typs ein Kriterium dafür, dass die Teilung
stark somatisiert gewesen ist — Aposporie, des Taraxacum-Typs dafür,
dass die Teilung weniger stark somatisiert — Semiaposporie, oder nur
unbedeutend somatisiert gewesen ist — Diplosporie. Die Fälle, wo dies
nicht stimmt, sind dadurch bedingt, dass die Gametophytogenese gleich-
zeitig einem Typ folgt, der dem Normaltyp nicht homolog ist.*

Gleichwie die Chromosomenform und die Bindung (vgl. FAGER-
LIND, 1940) als von dem Somatisierungsgrad der Teilung abhängig
angesehen werden können, so ist also auch die Wandbildung abhängig
von demselben. Sind die Ansichten richtig, so müssen in Sektion II
und III (Tabelle 2) Variationen, die auf stärkere meiotische Tendenz
deuten, gewöhnlicher sein als in Sektion I. Um diese Frage zu ent-
scheiden, bedarf es eingehenderer Untersuchungen, als sie bisher vor-
liegen. Ein Blick auf die Tabelle lehrt indessen, dass die Tendenz die
oben vermutete ist.

Die Tabelle zeigt, dass die meisten der Agamogonen nicht an einen
bestimmten Gametophytogenese-Typ gebunden sind. Variationen kom-
men vor. Die einzelnen Fälle unterscheiden sich offenbar hauptsäch-
lich voneinander durch die verschiedene Lage, die der Maximumpunkt
auf der Variationskurve einnimmt. Bei *Elatostema* und *Pellionia* spp.,
die ich eingehend untersucht habe (Resultate noch nicht veröffent-
licht), ist das Maximum deutlich weit nach der somatischen Seite hin
verschoben. Eine Variation gibt sich daher beim Studium dieser Arten
überhaupt nicht zu erkennen — wenigstens nicht, solange nicht ein
enorm grosses Material untersucht worden ist. Hierfür spricht auch,
dass bei nahestehenden Arten eine E.M.Z. oft nicht ausdifferenziert ist
(hier hat die Somatisierung ihr Maximum erreicht). Die Angabe, dass
bei *Elatostema acuminata* Variation vorkommt (TREUB, 1906; STRAS-
BURGER, 1910), beruht auf einem Irrtum. Dass dort viele Embryosack-
initialen vorhanden sind, erklärt sich daraus, dass somatische Aposporie
vorliegt (unveröffentlichte eigene Studien). Bei *Antennaria* und *Hiera-
cium* ist das Maximum mehr nach der meiotischen Seite hin verschoben
— die Variation wird reicher. Wenn schliesslich die Verschiebung in
dieser Richtung weiter geht, müssen die Variationen wieder beginnen
seltener zu werden. Einen solchen Fall bildet möglicherweise *Balano-
phora japonica*.

Nachdem die obige Arbeit niedergeschrieben war, hat GUSTAFSSON
(1939 b) eine neue Abhandlung veröffentlicht, in der die von mir er-
örterten Terminologiefragen Gegenstand der Behandlung gewesen sind.

GUSTAFSSON verwendet hier eine Terminologie, die teilweise von der früher von ihm benutzten abweicht. Sie weist einige Ähnlichkeiten mit der von mir vorgeschlagenen auf.

ZUSAMMENFASSUNG.

1. Unter Apomixis (WINKLER, 1908) versteht man Reproduktion, ohne dass Befruchtung und Kernphasenwechsel erfolgt sind.

2. Unter Agamospermie (TÄCKHOLM, 1922) versteht man Reproduktion mittelst Samen, ohne dass Befruchtung und Kernphasenwechsel erfolgt sind.

3. Für den Spezialfall von Agamospermie, wo die Reproduktion des Sporophyten über einen unreduzierten Gametophyten hin geschieht, wird der Terminus Agamogonie eingeführt.

4. Unter Parthenogenesis versteht man die Entwicklung der Eizelle zu einem neuen Individuum, ohne dass Befruchtung erfolgt ist, demnach nicht die ganze Entwicklungsreihe: Bildung eines unreduzierten Gametophyten aus einer Archesporzelle, Entwicklung des Gametophyten, Eizellbildung und weitere Entwicklung der Eizelle ohne Befruchtung.

5. Unter Apomeiosis (RENNER, 1916) versteht man die Bildung eines Gametophyten, ohne dass eine Reduktion der Chromosomenzahl stattgefunden hat.

6. Unter Diplosporie versteht man den Spezialfall der Apomeiosis, wo die erste Teilung der Initialzelle zur Entstehung unreduzierter Derivate geführt hat, die als Sporen angesehen werden können. Aus praktischen Gründen muss diese Forderung als erfüllt angesehen werden, wenn die erste Teilung stark meiotischen Charakters ist.

7. Unter Aposporie versteht man den Spezialfall von apomeiotischer Makrogametophytenbildung, wo die erste Teilung der Mutterzelle stark mitotischen Charakters ist. Ist die Mutterzelle eine Archesporzelle, so ist die Aposporie generativ; ist sie eine rein somatische Zelle, so ist die Aposporie somatisch. Die beiden Fälle generative und somatische Aposporie gehen ohne Grenze ineinander über.

8. Für den Fall apomeiotischer Makrogametophytenbildung, wo die erste Teilung der Mutterzelle ein Mittelding zwischen meiotischer und mitotischer Teilung darstellt, wird der Terminus Semiaposporie eingeführt.

9. Die Erscheinungen Diplosporie, Semiaposporie und Aposporie gehen ohne Grenze ineinander über, eine ganze Kette von Zwischenformen sind vielleicht vorhanden.

10. Diejenigen Fälle, wo bei apomeiotischer Makrogametophytenbildung die erste Teilung der Mutterzelle als eine mehr oder weniger asyndetische Meiosis, begleitet von Restitutionskernbildung, zu betrachten ist, werden aus praktischen Gründen als Diplosporie auch dann rubriziert, wenn die Asyndese als Kriterium beginnender Somatisierung anzusehen ist.

11. Wenn bei apomeiotischer Makrogametophytenbildung die erste Teilung der Mutterzelle eine pseudohomotypische Teilung ist, liegt Semiaposporie vor, da dieser Teilungstyp als eine Zwischenform von Meiosis und Mitose betrachtet wird.

12. Wenn bei apomeiotischer Makrogametophytenbildung die Mutterzelle eine Archesporzelle ist und bei ihrer ersten Teilung die Chromosomen hochgradig somatische Form aufweisen (*Eupatorium*-Typ, *Hieracium*-Typ), liegt generative Aposporie vor.

13. Wenn generative Aposporie vorliegt, folgt die Embryosackentwicklung stets dem *Antennaria*-Typ.

14. Wenn Diplosporie oder Semiaposporie vorliegt, folgt die Embryosackentwicklung, sofern sie auf eine dem Normaltyp homologe Weise geschieht, stets dem *Taraxacum*-Typ.

15. Wenn Diplosporie oder Semiaposporie vorliegt, folgt die Embryosackentwicklung, sofern sie auf eine dem bisporischen oder dem tetrasporischen Typ homologe Weise geschieht, stets dem *Antennaria*-Typ.

Botanisches Institut der Universität Stockholm, im Juli 1939.

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ZYTOLOGIE UND GAMETOPHYTENBILDUNG IN DER GATTUNG WIKSTROEMIA

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EINLEITUNG.

WÄHREND seines Aufenthalts im Botanischen Garten zu Buitenzorg auf Java fiel es HANS WINKLER (1904, 1906) auf, dass dort kultivierte Individuen von *Wikstroemia indica* trotz abnormen, in hohem Grade abortiven Pollens Früchte und Samen bildeten, die, wie es sich zeigte, auch Embryonen enthielten. Er vermutete, dass es sich um einen Fall von apomiktischer Samenbildung handelte, was ihm auch durch Kastrierversuche völlig bindend zu beweisen gelang. Bei der Pflanze kam es nur zur Bildung weniger Samen. Diese wurden in der Regel nur von den mehr basalen Blüten der Blütenstände erzeugt. Wenn in einem Blütenstand alle Blüten mit Ausnahme einiger basalen frühzeitig beseitigt wurden, erhielt man unter im übrigen normalen Verhältnissen eine Frucht- und Embryobildung von 39,1 %, was bedeutend mehr ist, als wenn die erwähnte Operation nicht vorgenommen worden wäre. In auf dieselbe Weise behandelten Blütenständen, wo aber die Blüten ausserdem kastriert worden waren — WINKLER begnügte sich nicht damit, die Staubgefässe zu entfernen, er schnitt auch die Narben weg — war die Frucht- und Embryobildung 34,7 %. Die Differenz zwischen diesem Resultat und dem vorigen war also verhältnismässig unbedeutend und dürfte wohl dem gewaltsamen Eingriff in die Blüte zuzuschreiben sein, den die Kastrierung darstellte. In P.M.Z. bestimmte WINKLER die Chromosomenzahl zu $n = 26$. Der Embryosack war seiner Meinung nach direkt aus der E.M.Z. gebildet. Eine Synapsis im Kern der letzteren wurde nie beobachtet. Er zeigte, dass der Embryo sich aus der Eizelle entwickelte. Einen Fall von Nucellarembryonie glaubte er gefunden zu haben (Fig. 31 in WINKLERS Arbeit).

Eine erneute Untersuchung von *Wikstroemia indica* — offenbar von demselben Klon, mit dem WINKLER gearbeitet hatte — wurde von STRASBURGER (1909) ausgeführt, dem es gelang, WINKLERS Schilderung in einigen Punkten zu ergänzen. Die E.M.Z. teilte sich zuerst in zwei Zellen, von denen eine den Embryosack bildete. »Eine feste Scheide-

wand wird zwischen den beiden Zellen nicht ausgebildet, sie erscheinen
vielmehr nur durch einen hellen Zwischenraum voneinander getrennt
(Fig. 43, 44, 45, Taf. II), so dass leicht bei geneigter Lage dieser Tren-
nungsfläche die Vorstellung eines einzigen, zweikernigen Protoplasten
hervorgerufen werden kann». Triaden und Tetraden wurden in einigen
wenigen Fällen von STRASBURGER beobachtet. Das Vorkommen von
Dyaden war von WINKLER nicht übersehen worden, er hielt sie jedoch
für Ausnahmeerscheinungen. STRASBURGER verwendete viel Arbeit auf
den Versuch, zur Klarheit über die Prozesse zu kommen, die während
der ersten Teilung in der E.M.Z. stattfanden. Synapsis und Diakinese
kamen nie vor. In einigen Fällen (einem Falle?) zeigte die E.M.Z. in
Prophase Chromosomen somatischen Aussehens (Fig. 38 b bei STRAS-
BURGER). Während der Meta- und der Anaphase traten Chromosomen
in einer Anzahl auf, die sich dem Wert 26 näherte. Sie hatten ein Aus-
sehen, das mit dem für Meiosis gewöhnlichen übereinstimmte. Trotz-
dem meinte STRASBURGER, dass die Teilung hier eine Äquationsteilung
sei. In einer späteren Arbeit findet STRASBURGER (1910) eine variierende
Anzahl somatischer Chromosomen (22—29). Seiner Ansicht nach stellt
hier in den meisten Fällen jedes somatische Chromosom in Wirklichkeit
zwei dar, die sich nicht voneinander geschieden haben. Den von
WINKLER vermeintlich beobachteten Fall von Nucellarembryonie will
er nicht anerkennen. Einige nahverwandte Arten wurden als Vergleichs-
objekte untersucht. *Daphne Mezereum, D. alpina, Wikstroemia ca-
nescens* und *Gnidia carinata* hatten alle n=9 (STRASBURGER, 1909, 1910).
Nichts wurde beobachtet, was auf anormale Reproduktion oder Gameto-
phytenbildung bei diesen deutete.

Um *Wikstroemia* einer erneuten Untersuchung zu unterziehen,
sammelte ich während eines Aufenthalts in Buitenzorg ein reichliches
Material von *Wikstroemia indica* ein, die subspontan in grosser Menge
dicht vor dem Garten vorkommt. Dieser Bestand stellt laut Angabe
seitens der Gartenleitung und auch im Herbarium in Buitenzorg die
Nachkommenschaft der nun ausgestorbenen, früher im Garten kulti-
vierten Individuen dar, eben der Individuen, die von WINKLER und
STRASBURGER studiert worden sind. Es kann also als sicher angesehen
werden, dass mein Material und das »klassische» von ein und dem-
selben Klon herstammen. Die ursprünglichen Individuen sind laut An-
gabe ursprünglich dem Garten in Buitenzorg von dem Botanischen
Garten in Calcutta überwiesen worden. STRASBURGER (1909, S. 87) ver-
folgte ihre Herkunft weiter bis nach Srirampur in Bengalen. *Wikstroe-
mia indica* ist eine polymorphe Art (STRASBURGER, GILG — vgl. STRAS-

BURGER, 1909, S. 85 und beispielsweise KOORDERS, 1911). Die in Buitenzorg wachsende gehört dem Formenkreis *viridiflora* MEISSN. an.

Während seines letzten Besuchs auf Hawaii fixierte Professor CARL SKOTTSBERG Exemplare einiger *Wikstroemia*-Arten, die er Professor OTTO ROSENBERG überliess. Als letzterer erfuhr, dass ich mit einer Untersuchung von *Wikstroemia indica* beschäftigt sei, hatte er die Freundlichkeit, das SKOTTSBERGsche Material mir zur Verfügung zu stellen. Die Überweisung dieses Materials, durch das ein sehr erwünschtes Vergleichsmaterial erhalten wurde, ist von grosser Bedeutung für meine Arbeit gewesen.

Sämtliche Fixierungen waren mit »Karpechenko» geschehen, eventuell mit Vorfixierung in Carnoy oder abs. Alkohol. Von den *indica*-Fixierungen erwiesen sich leider mehrere als völlig unbrauchbar. Ein geringerer Teil war dagegen in sehr gutem Zustand. Das Material genügte zur Lösung einer Reihe von Fragen.

Unten wird die von mir (FAGERLIND, 1940) vorgeschlagene Terminologie benutzt.

DIE SPOROGENESE BEI AMPHIMIKTISCHEN WIKSTROEMIA-ARTEN.

Das hawaiische Material bestand aus fünf verschiedenen Arten: *W. phillyraefolia* A. GRAY, *pulcherrima* SKOTTSB., Nr. 2726 (steht nach SKOTTSBERG *W. furcata* (HBD.) nahe), Nr. 2737 (ist nach SKOTTSBERG wahrscheinlich *W. uva ursi* A. GRAY) und 16/8—38 (nach SKOTTSBERG eine vielleicht endemische Art von der Insel Kauai). Alle diese Arten erwiesen sich als diözisch. Männliche Blüten waren erhalten worden von 2737, 16/8—38 und *pulcherrima*, weibliche Blüten von *phillyraefolia*, 2726 und *pulcherrima*.

Auch in den weiblichen Blüten werden Staubgefässe ausgebildet, in denen P.M.Z. sich herausdifferenzieren. Die letzteren machen eine normale Meiosis durch. Bevor die Tetraden sich aufgelöst haben, degeneriert indessen die ganze »Tetradenmasse». Die Meiosis in den P.M.Z. verlief normal bei sämtlichen Arten des hawaiischen Materials. Bei 2726 und 2737 hat die Chromosomenzahl zu $n = 9$ bestimmt werden können. Die anderen drei Arten sind wohl auch diploid $(n = 9?)$, die Zahl in somatischen Zellen, die in Teilung begriffen waren, konnte bei ihnen allen als etwa 18 bestimmt werden. Die Pollenbildung folgt dem simultanen Schema. Eine temporäre Zellplatte fand sich jedoch stets gleich nach dem Abschluss der ersten Teilung. Pollenkörner mit zwei und ältere mit drei Kernen sind beobachtet worden.

In den weiblichen Blüten waren Stempel und Samenanlagen in der Weise aufgebaut, wie WINKLER und STRASBURGER es geschildert haben. Das primäre Archespor bildet Deckzelle und E.M.Z. Die erstere sowie die Nucellusepidermis teilen sich, wodurch die Nucellusmasse zunimmt. Die E.M.Z. ist in der Regel in Einzahl vorhanden, vereinzelt können zwei solche beobachtet werden (Fig. 6—7). E.M.Z. mit dem Kern in Synapsis werden oft angetroffen. Das Resultat der Meiosis ist eine Zellentetrade, oft ausgebildet zu einer T-Tetrade (Fig. 8). Die Basalzelle bildet in der Regel den Embryosack (Fig. 9). In dem reifen
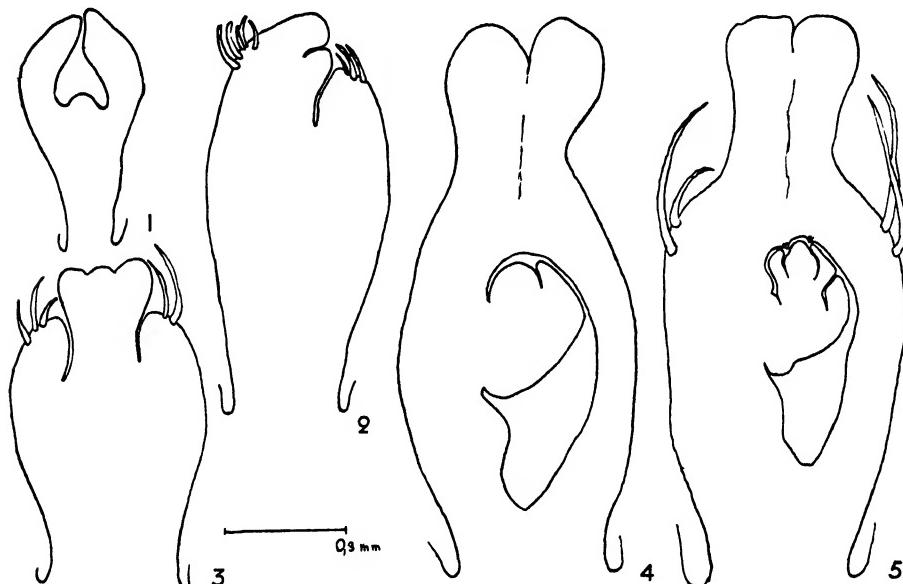
Fig. 1—4. Rudimentäre Stempel von männlichen Blüten verschiedener amphimiktischer *Wikstroemia*-Arten. — Fig. 5. Normaler Stempel bei *Wikstroemia viridiflora*.

8-kernigen Embryosack degenerieren bald die kleinen Antipoden. Die Polkerne verschmelzen nahe der Basis des Embryosacks. Eine Synergide wird oft in degenerativem Zustand beobachtet, eine Erscheinung, der man ja stets begegnet, gleich nachdem der Pollenschlauch eingedrungen ist. Die hawaiischen *Wikstroemia*-Arten, von denen weibliche Blüten mir zur Verfügung gestanden haben, sind demnach sicher Amphimikten.

Wie die weiblichen Blüten Staubgefässe enthalten, in denen doch keine funktionierenden Sporen gebildet werden, so enthalten die männlichen Blüten mehr oder minder abnorme Stempel. Diese sind bei *phillyraefolia* sehr reduziert (Fig. 1). Bei 16/8—38 sind sie äusserlich

ziemlich normal entwickelt (Fig. 4). Die Samenanlage ist jedoch nur mit einem Integument versehen (2 sind das Normale), mit einem Integument, das nicht einmal vollständig den Nucellus umhüllt und eines Archespors völlig entbehrt. Bei *pulcherrima* findet man meistens die eigentümlichen Stempel, die in Fig. 2 und 3 wiedergegeben sind. In Fig. 2 ist eine Samenanlage kaum ausgebildet, in Fig. 3 bildet sie eine
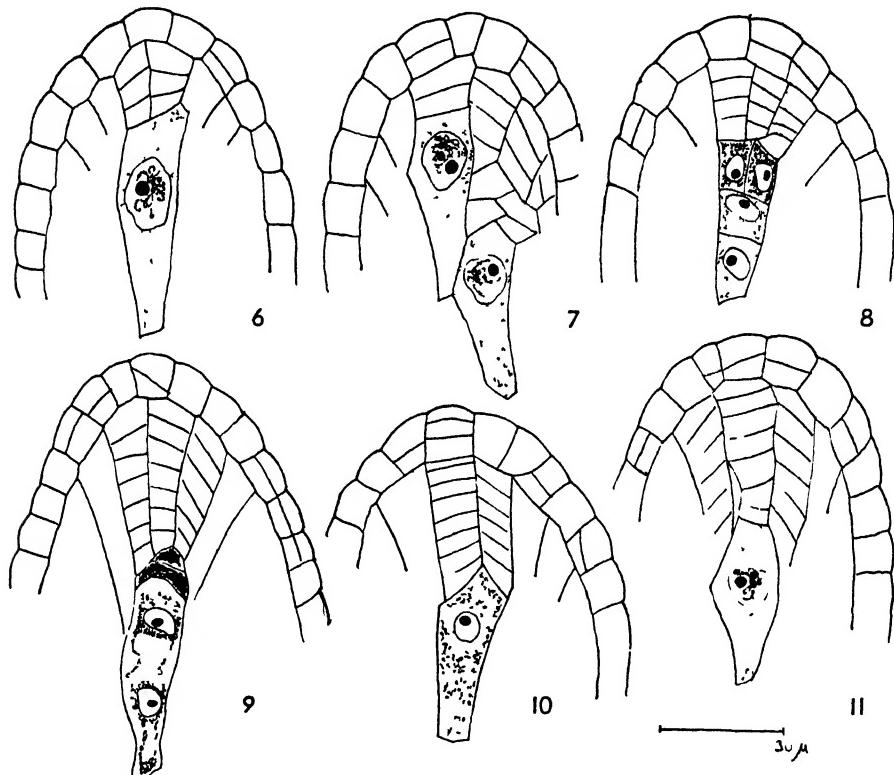

Fig. 6—9. Amphimiktische *Wikstroemia*-Arten. — 6. Einzelliges Archespor. — 7. Zweizelliges Archespor. — 8. Tetrade. — 9. Zweikerniger Embryosack. — Fig. 10—11. *Wikstroemia viridiflora*. — 10. E.M.Z. mit Kern in Ruhe. — 11. E.M.Z. mit Kern in Synapsis.

direkte Fortsetzung der Zentralachse des Stempels, ragt aus der Spitze des Stempels heraus. Wenn sie atrop und mit einem symmetrisch ausgebildeten Integument versehen ist, erhält man den Eindruck, dass die Blütenachse verlängert und mit zwei Kränzen aus zusammengewachsenen Blättern versehen ist. Diese eigentümlichen Stempel werden zuweilen auch bei den weiblichen Blüten anderer *Wikstroemia*-Arten angetroffen. Mehrere Fälle von eigentümlich ausgebildeten Stempeln

bei männlichen Individuen gewisser *Wikstroemia*-Arten werden von
SKOTTSBERG (1936) abgebildet.

DIE MIKROSPOROGENESE BEI EINEM APOMIKTISCHEN KLON VON WIKSTROEMIA INDICA.

Angaben über die Entwicklung des männlichen Archespors und
des Tapetumgewebes bei *Wikstroemia indica* finden sich bei WINKLER,
weshalb von einer Schilderung derselben hier abgesehen werden kann.
Während die P.M.Z. sich in Prophase befinden, erfolgt oft Cytomixis.
Zuweilen wird eine wirkliche Verschmelzung von P.M.Z. und ihren
Kernen beobachtet. Die Prophase hat in verschiedenen Kernen ver-
schiedenes Aussehen, was dann natürlich auf das der späteren Phasen
einwirkt. Die Einzelheiten im Verlauf der Prophase haben leider nicht
studiert werden können, da die Beschaffenheit des Materials derart ist,
dass distinkte Bilder davon nicht erhalten werden können. Einige
P.M.Z.-Kerne erinnern sehr an somatische. Diese werden unten ein-
gehender behandelt werden. Diejenigen P.M.Z.-Kerne, die, dem Aus-
sehen nach zu urteilen, die früheren meiotischen Prophasestadien durch-
gemacht haben, gehen allmählich in Diakinese über. Die Diakinese-
kerne sind hier von recht verschiedenem Aussehen, sie sind jedoch stets
durch hochgradige Asyndese gekennzeichnet. In nicht wenigen Fällen
sind Gemini in wechselnder Anzahl, zwischen 9 und 1, zu beobachten
(Fig. 12 und 13). Die Univalente variieren in diesen Fällen zwischen
9 und 25. Der studierte *Wikstroemia indica*-(*viridiflora*-)Klon ist also
triploid (vgl. die oben behandelten *Wikstroemia*-Arten). Die Pollenbil-
dung in diesen Fällen stimmt demnach mit dem überein, was ROSENBERG
(1917, 1927) in seinen bedeutungsvollen *Hieracium*-Untersuchungen als
boreale-Typ bezeichnete. In anderen Fällen, die ebenfalls gewöhnlich
sind, ist die Asyndese vollständig, es treten 27 Univalente auf. Dieser
Fall entspricht innerhalb der *Hieracium*-Literatur dem *laevigatum*-Typ.
Sind Gemini vorhanden, so werden diese in einen Äquator eingeordnet.
In einem Falle ist in einem solchen Äquator eine Bildung angetroffen
worden, die als ein Trivalent gedeutet werden kann (Fig. 13). Mög-
licherweise handelt es sich um ein Bivalent, mit dem ein Univalent in
Kontakt gebracht worden ist. Die Univalente liegen zerstreut in der
Spindel (Fig. 13—14), die Teilung ist also ihrem Charakter nach semi-
heterotypisch (ROSENBERG, 1927). Die Gemini teilen sich, und die Uni-
valente verteilen sich nach dem Zufallsgesetz. Nicht wenige Univalente
ordnen sich jedoch bisweilen in den Äquator ein (Fig. 16). Diese und

auch andere, zufällig zentraler gelegene Univalente zeigen oft eine Neigung zu beginnender Teilung (Fig. 18), die bisweilen auch durchgeführt
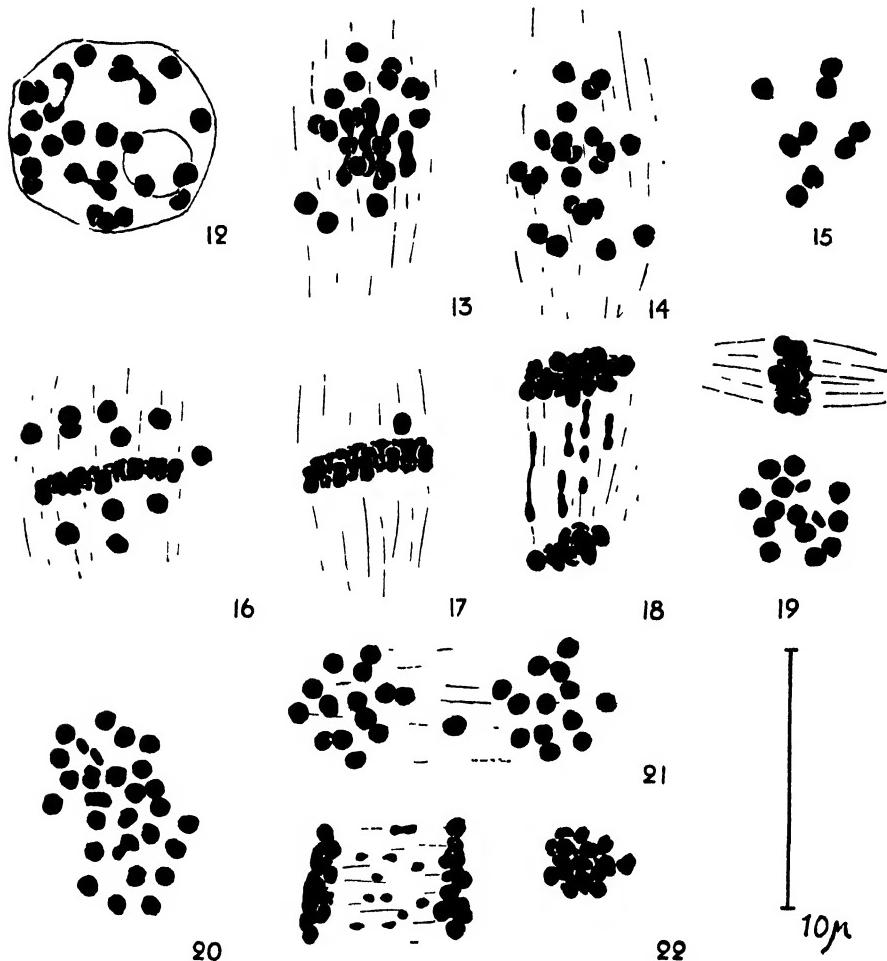

Fig. 12—22. *Wikstroemia viridiflora* — ›normale Meiosis‹ in der P.M.Z. — 12. Diakinese mit $3_{II} + 21_I$. — 13. Metaphase I mit 1_{III} (?) $+ 4_{II} + 16_I$. — 14. Semiheterotypische Teilung nur mit Univalenten (Teil eines Kerns). — 15. Teil eines solchen Kerns mit starker ›sekundärer Assoziation‹. — 16. Teilung I, mehrere Univalente sind in den Äquator eingewandert. — 17. Alle Univalente bis auf einen sind in den Äquator eingewandert. — 18. Anaphase I mit Nachzüglern, die Teilungstendenz zeigen. — 19. Metaphase II mit $13_I + 2_I^1$. — 20. Metaphase II nach Restitutionskernbildung mit $26_I + 2_{\frac{1}{2}}^1$. — 21. Anaphase II mit starker ›sekundärer Assoziation‹. — 22. Anaphase II mit Nachzüglern.

wird. Im letzteren Falle werden also Hemiunivalente erzeugt. Sie sind deutlich an ihrer geringen Grösse zu erkennen (Fig. 19—20). Selten

geschieht es, dass sämtliche Univalente in einen Äquator eingeordnet
sind (vgl. Fig. 17). Die folgende Teilung geschieht in solchem Falle
natürlich äquational. Dieser Teilungstyp ist identisch mit dem, was
GUSTAFSSON (1934 und später) pseudohomotypische Teilung genannt
hat. In vielen Fällen, wo die Teilung in dieser Weise verlaufen ist,
haben die Chromosomen in geringerem Grade die kontrahierte Gestalt,
die für die Meiosis charakteristisch ist. Hierüber mehr unten.

Während der Anaphase der ersten Teilung entstehen meistens zwei
Chromosomengruppen, eine an jedem der beiden Pole, und zwischen
ihnen finden sich ungeteilte oder unvollständig geteilte Nachzügler. In
der Regel dürften die letzteren sich jedoch noch in die betreffenden Pol-
gruppen einordnen, bevor die Kernmembranen gebildet werden. In
anderen Fällen werden Restitutionskerne oder zwischen den »legitimen»
Dyadenkernen Mikrokerne gebildet. Während der Interkinese ist nicht
selten eine Membranbildung im Phragmoblasten zu beobachten. Sie
scheint sehr temporärer Natur zu sein, und nie kommt es zu einer wirk-
lichen Wandbildung. Metaphase II zeigt infolge der früheren Unregel-
mässigkeiten verschiedene Chromosomenzahlen. Ausser Chromosomen
von »normaler» Grösse bemerkt man einige »Kleinchromosomen»,
sicherlich identisch mit den Hemiunivalenten (Fig. 19). Während der
Anaphase II sieht man gleichfalls Nachzügler, die klein sind und als
mit den Hemiunivalenten identisch betrachtet werden können (Fig. 22).
Wenn ein Restitutionskern durch die erste Teilung zustande gekommen
ist, kommt während der zweiten nur eine einzige grosse Chromosomen-
gruppe vor, die sich dann teilt (Fig. 20). In dieser können auch oft die
Hemiunivalente beobachtet werden. Die Anzahl Chromosomenkörper
in einer derartigen unreduzierten Chromosomenplatte kann daher
grösser als 27 sein. Rechnet man jeden der kleinen Körper als ein
halbes Chromosom, so erhält man jedoch ein übereinstimmendes Resul-
tat. In Fig. 20 finden sich so 2 »Kleinchromosomen» und 26 Chromo-
somen von gewöhnlicher Grösse.

War die Entwicklung in der Weise gegangen, wie oben geschildert
worden, so hatten die Univalente während der Diakinese stark kontra-
hierte Form, sie ähnelten Kugeln oder Würfeln. Diakinesen — wenn
nun weiter diese Bezeichnung angewandt werden darf — anderen Aus-
sehens werden aber bisweilen angetroffen. Bald haben die Chromosomen
ovale Gestalt, bald haben sie die Form kurzer, dicker, an der Mitte gebo-
gener Stäbchen (Fig. 25), bald sind sie stark in die Länge gezogen (Fig.
26). Im letzteren Falle unterscheiden sie sich nicht von dem Aus-
sehen, das den Chromosomen während der Prophasen zu den Tei-

lungen zukommt, welche im somatischen Gewebe zu beobachten sind. Ein Gegenstück hierzu bei *Hieracium* bildet der *pseudoillyricum*-Typ
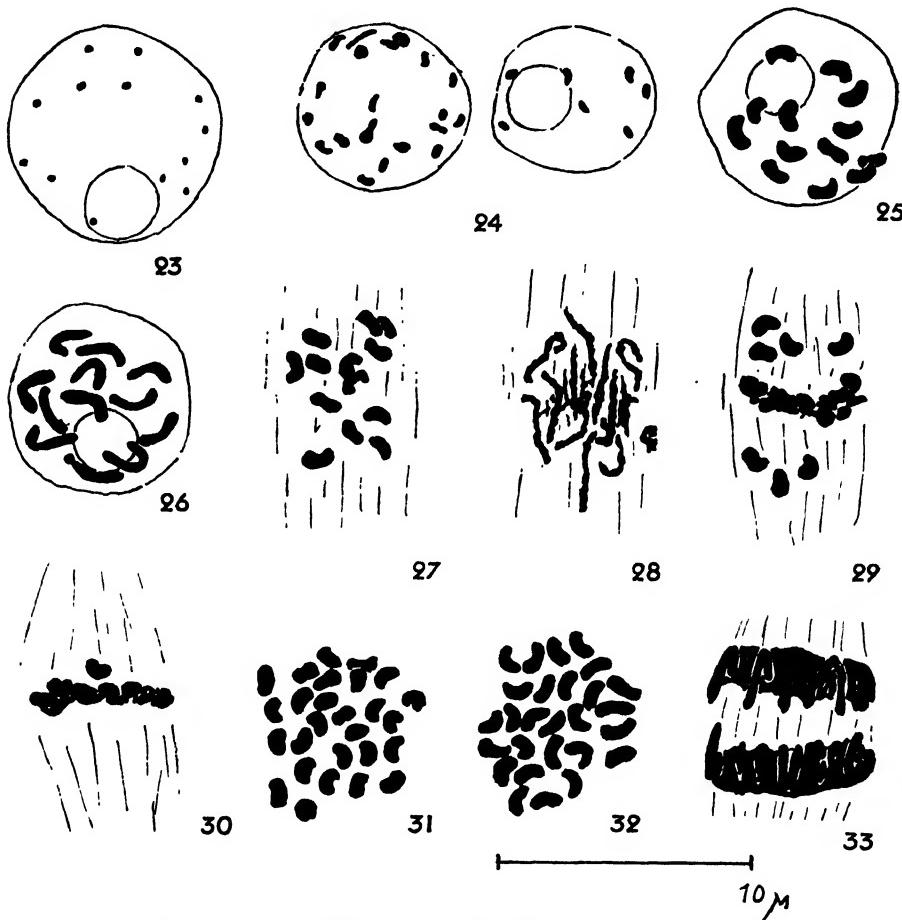

Fig. 23—33. *Wikstroemia viridiflora* — ›somatisierte Teilungen› in der P.M.Z. — 23. Prophase mit Prochromosomen (?). — 24. Desgl., etwas älteres Stadium. — 25. ›Diakinese› nur mit Univalenten von ›schwach somatisierter Form›. — 26. ›Diakinese› nur mit Univalenten von ›stark somatisierter Form›. — 27. ›Schwach somatisierte Univalente› ordnen sich in den Äquator ein. — 28. ›Stark somatisierte Univalente› ordnen sich in den Äquator ein. (Die Kerne in Fig. 27 und 28 stammen aus in demselben Fach nebeneinander liegenden P.M.Z.) — 29—30. Die ›Univalente von schwach somatisierter Form› sind sämtlich oder zum grössten Teil in den Äquator eingewandert. — 31—32. ›Univalente von schwach und mittelstark somatisierter Form› haben ›pseudohomotypische Metaphase› gebildet. — 33. Stark somatisierte Chromosomen während Anaphase I.

(ROSENBERG, 1927). Im erstgenannten Falle kommt dann bisweilen eine semiheterotypisch betonte Teilung zustande. Während dieser ist jedoch

die Tendenz zur Einverleibung von Univalenten in den Äquator stärker
(Fig. 29—30) als während des »normalen», oben geschilderten Ver-
laufs. In vielen Fällen ordnen sich auch sämtliche Chromosomen zu
einer völlig regelmässigen Äquatorialplatte an. Das Resultat ist dann
die pseudohomotypische Teilung oder besser gesagt, wenn man sich an
die Chromosomenlänge hält, eine Zwischenform zwischen diesem
Teilungstyp und einem somatischen. Waren die Chromosomen nach
Alternative II ausgebildet, so war dieser Teilungstyp noch gewöhnlicher
und zwar der alleinherrschende, wenn die Chromosomen somatische
Gestalt hatten. Aus dem Angeführten geht hervor, dass die Kerne in
den P.M.Z., wenn man sich an die Chromosomenform hält, »somati-
siert» werden können. Alle Übergänge zwischen der Bildung von Uni-
valenten »meiotischen» und solchen »mitotischen» Aussehens sind dem-
nach vorhanden. Wenn die mitotische Form sich etwas geltend gemacht
hat, fehlen stets Gemini. Wie die Kerne, die sich auf die soeben be-
schriebene Weise verhalten, später sich entwickeln, ist mir unbekannt.
Die Kerne, die sich in der Richtung auf »Somatisierung» hin entwickelt
haben, zeigen keinerlei Verzögerung in ihrer Entwicklung. Ein Parallel-
fall zu dem eben geschilderten, wo die P.M.Z. bei ein und derselben
Art sich sowohl nach dem *boreale-* als auch nach dem *laevigatum-* und
dem *pseudoillyricum*-Typ entwickeln können, ist von GENTSCHEFF
(1937) bei *Hieracium vulgatum* nachgewiesen worden. Schon ROSEN-
BERG zeigte übrigens, dass mehr als einer der drei Entwicklungstypen
bei ein und derselben Art angetroffen werden können.

Wie oben erwähnt, bieten einige P.M.Z. während der Prophase ein
Aussehen dar, das nicht auf beginnende Meiosis deutet. Vermutlich
sind es diese Fälle, bei denen es zu den in ausgesprochenem Grade
»somatisierten» Teilungen kommt. Statt der zu erwartenden meioti-
schen Prophasen haben die Kerne in einigen Fällen das Aussehen, das
durch Fig. 23—24 veranschaulicht wird. In dem Kern finden sich hier
ausser dem Nukleolus kleinere oder grössere distinkte Punkte. Ihre
Anzahl ist 27. Sie sind wohl als Prochromosomen aufzufassen, als
Chromozentren, um die herum die Chromosomen später »aufgebaut»
werden (vgl. ROSENBERG, 1909 b; DOUTRELIGNE, 1933). In einem
Pollenfach sind in einer Reihe liegende Kerne in den Stadien beobachtet
worden, die durch Fig. 23, 24 und 25 veranschaulicht werden; es zeigt
dies, dass sie Phasen einer und derselben Entwicklungskette darstellen.

Eine ganze Reihe P.M.Z. scheinen überhaupt keine Teilungen
durchzumachen. Sie nehmen dann oft ein Aussehen an, identisch mit
dem der Tapetumzellen. Auf diese Weise werden die Pollenfächer oft

regelmässig in Kammern abgeteilt. Nicht selten schwellen diese »ruhenden» P.M.Z. abnorm an, eine Erscheinung, die bisweilen auch bei den Zellen des Tapetumgewebes zu finden ist.

Der »reife» Pollen bleibt stets einkernig. Er ist von sehr verschiedener Grösse und Form. Dass er letal ist, dürfte aus WINKLERS Keimversuchen hervorgehen. WINKLER, der eine Menge frischer Pollenfächer auf ihren Inhalt analysiert hat, gibt an, dass in ganz vereinzelten Fällen ein Fach mit gleichförmigem Pollen vitalen Aussehens angefüllt sein kann. Einen solchen Fall habe ich nicht zu entdecken vermocht.

Während der meiotischen Teilungen wird oft sog. sekundäre Assoziation beobachtet. Die Assoziationskomplexe bestehen zumeist aus 2 Chromosomen (Fig. 14, 15, 19, 20, 21). Dreiergruppen sind nicht gewöhnlich. Grössere Gruppen, die jedoch mehr den Eindruck von »Zusammenballung» machen, kommen zuweilen vor. Assoziationen kommen auch zwischen den Univalenten vor, die in der Spindel zerstreut liegen (Fig. 14, 15). Wären die Assoziationen ein Kriterium für Homologie, so müssten Dreiergruppen gewöhnlich sein. Dies ist jedoch nicht der Fall. Dass die Assoziationen in der semiheterotypischen Spindel als Pseudogemini zu betrachten sind, in dem Sinne, wie GUSTAFSSON diesen Ausdruck anwendet, und dass die Assoziationen wirklich ein Homologiekriterium darstellen, möchte ich andauernd bezweifeln (vgl. FAGERLIND, 1937).

Bei *Galium* beschrieb ich (FAGERLIND, 1937) Fälle, wo während der Anaphase, eben wenn die Chromosomenmassen die betreffenden Pole erreicht hatten, die Assoziation nicht dieselbe in korrespondierenden Platten war. Ähnliche Fälle können auch hier beobachtet werden; Fig. 21 ist ein solcher.

DIE »MAKROSPOROGENESE» BEI EINEM APOMIKTISCHEN KLON VON WIKSTROEMIA INDICA.

Im Nucellus teilt sich die primäre Archesporzelle in Deckzelle und E.M.Z., wie WINKLER dies beschrieben hat. Die Deckzelle nebst Epidermis teilt sich mehrmals, wodurch die Nucellusmasse zunimmt (Fig. 10, 11). Bisweilen können zwei E.M.Z. in demselben Nucellus angetroffen werden (Fig. 34, 36). Meistens werden von der E.M.Z nur zwei Zellen gebildet (Fig. 35—37, 39, 40). Seltener ist Tetradenbildung (Fig. 38). Der Verlauf bei der Megagametophytenbildung stimmt also im grossen ganzen mit den von STRASBURGER gelieferten Angaben überein. Die Zellen sind indessen voneinander durch wirkliche Wände

geschieden, nicht nur durch einen »hellen Zwischenraum». Die gebil-
deten Zellen liegen zuweilen direkt übereinander. Oft sind sie etwas
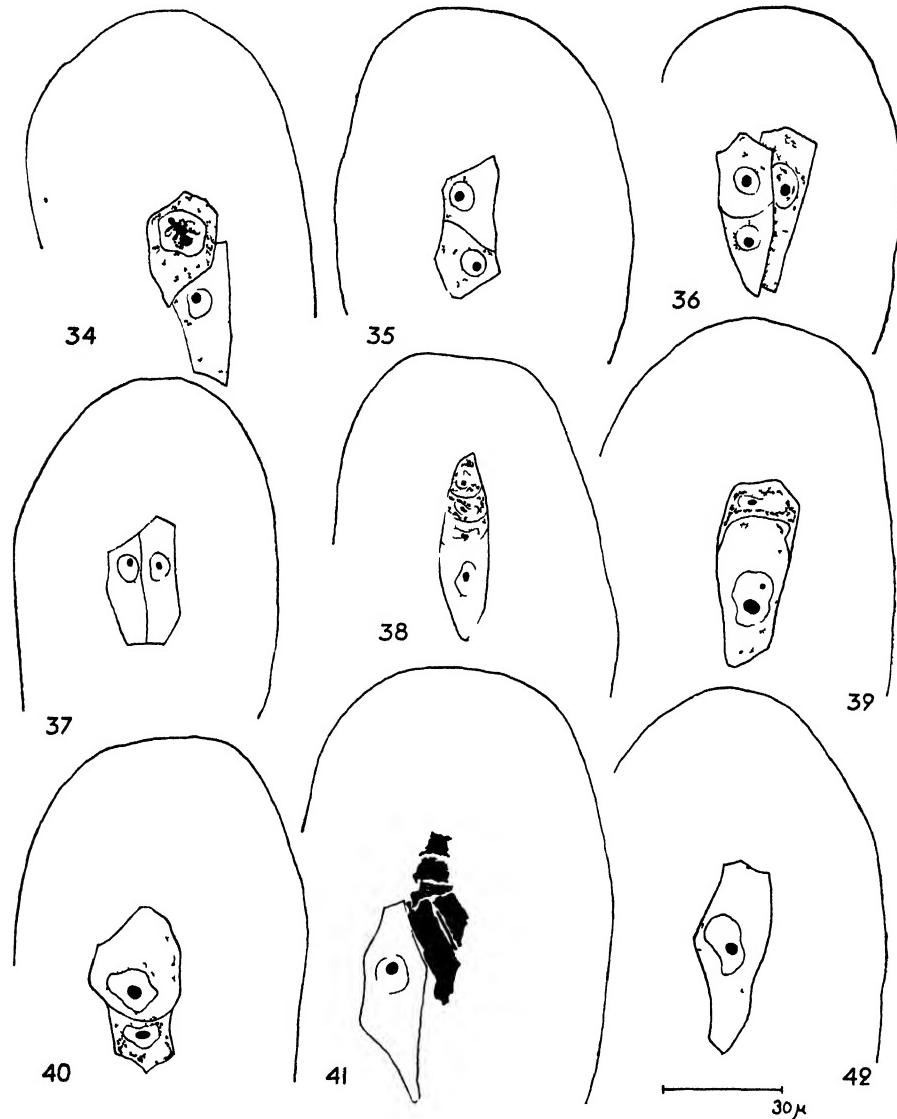


Fig 34—42 *Wikstroemia viridiflora.* — 34 Zweizelliges Archespor, eine E M Z mit
Kern in Synapsis, die andere mit Ruhekern. — 35 Dyade — 36 Dyade und un-
geteilte E M Z mit Kern in Ruhe. — 37. Dyade mit den Tochterzellen in Scitenlage.
— 38. Tetrade — 39—40. Junge Embryosacke, gebildet nach dem *Taraxacum*-
Schema. —·41. Degenerierte Tetrade (?) und junger Embryosack, gebildet nach dem
Antennaria-Schema. (?). — 42. Junger Embryosack, gebildet nach dem *Antennaria*-
Schema (?).

verschoben im Verhältnis zueinander. Liegt dann die Schnittebene in bestimmter Weise, so ist es schwer, die Wände zu sehen. Hierin liegt wohl die Erklärung zu STRASBURGERS irrtümlicher Angabe. Sehr selten liegen die Tochterzellen nebeneinander (Fig. 37), ein Fall, der auch von STRASBURGER beobachtet worden ist. Dass es sich hierbei nicht um zwei nebeneinander liegende E.M.Z. handelt, geht völlig deutlich aus dem Aussehen der Kerne und des Plasmas hervor. Die beiden Zellen in der Dyade zeigen Entwicklungstendenz. Beide können vakuolisiert werden. In der Regel verdrängt jedoch die untere die obere, die sogleich degeneriert (Fig. 39). In vereinzelten Fällen kann der Verlauf der gerade entgegengesetzte sein (Fig. 40). Einige Male habe ich den Eindruck gehabt, dass die Bildung eines Embryosacks direkt aus einer E.M.Z. erfolgte, ohne dass diese sich zuerst geteilt hatte (Fig. 41—42). E.M.Z. mit Ruhekernen haben nämlich beginnende Vakuolisierung aufgewiesen. In späteren Stadien zu entscheiden, ob der Embryosack aus einer Dyadenzelle oder aus einer ungeteilten E.M.Z. hervorgegangen ist, stellt sich schwierig, da mehrere somatische Zellen im Nucellus gleichzeitig mit der eventuellen Schwesterzelle degenerieren und verdrängt werden. Es ist daher unmöglich, einen Anhaltspunkt durch Bestimmung der Anzahl degenerierter Zellreste um die Basis oder Spitze des jungen Embryosacks herum zu erhalten.

Bei dem studierten agamogonischen Klon von *Wikstroemia viridiflora* folgt also die Embryosackbildung hauptsächlich dem sog. *Taraxacum*-Schema.

Leider ist das Material so gering gewesen, dass E.M.Z. in Teilung nur einige wenige Male haben beobachtet werden können. Untersucht man Nucelli, die, dem Aussehen nach zu urteilen, sich in dem Stadium befinden, wo der E.M.Z.-Kern bei den sexuellen *Wikstroemia*-Arten in Synapsis begriffen ist, so trifft man nur ruhende E.M.Z.-Kerne an. Später können jedoch Kerne in Synapsis angetroffen werden, aber nur selten (Fig. 11, 34). Zwei Fälle von Diakinese sind beobachtet worden. Beide zeigen Asyndese, aber auch Bindung. Fig. 43 zeigt vermutlich sogar ein Trivalent. Fig. 45 zeigt eine Metaphase I, wo die Chromosomen univalent und von meiotischer Form sind. Die meisten sind in den Äquator eingeordnet worden, nur zwei befinden sich ausserhalb desselben. In Fig. 46, die den zweiten angetroffenen Fall von Metaphase I wiedergibt, sind sämtliche Univalente in den Äquator eingeordnet. Sie haben meiotische Gestalt. Die Teilung ist offenbar pseudohomotypisch. Die Embryosackbildung kann also durch Semiaposporie erfolgen. Fig. 47 zeigt, dass während der Anaphase I Nachzügler vor-

kommen können. In einigen Fällen, wo der Kern sich in Synapsis befunden hat, hat eine Weiterentwicklung offenbar nicht geschehen können. Der E.M.Z.-Kern hat lange dieses sein Aussehen beibehalten. Die Zelle und der Kern degenerieren ohne weitere Veränderungen. Die Nucelli bleiben in solchen Fällen steril.

Viele Fälle finden sich, wo die E.M.Z., trotzdem ein spätes Stadium vorliegt (Fig. 10, 34, 36), nie Synapsisaussehen zeigt, sondern in Ruhe
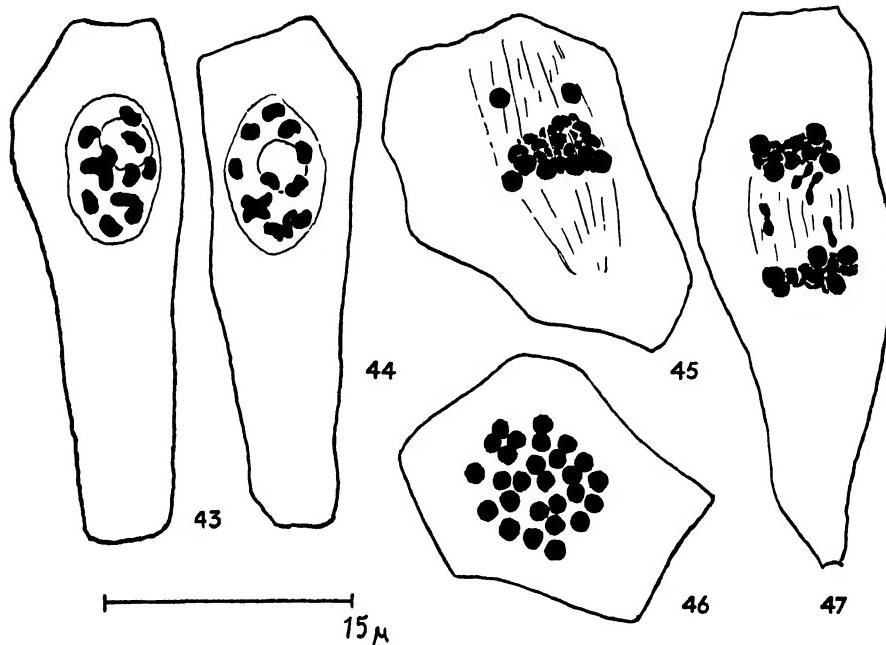

Fig. 43—47. *Wikstroemia viridiflora* — die erste Teilung des E.M.Z.-Kerns. — 43. Diakinese mit Trivalent (?), Geminus und Univalenten (Kernfragmenten). — 44. Desgl. mit Gemini und Univalenten. — 45. Metaphase I nur mit Univalenten, von denen alle bis auf zwei in den Äquator eingewandert sind. — 46. Pseudohomotypische Metaphase. — 47. Anaphase I mit Nachzüglern.

bleibt. Es muss dies teilweise darauf beruhen, dass das Synapsisstadium sehr rasch vorübergeht. Dies ist auch wohl die Ursache, weshalb WINKLER und STRASBURGER die Existenz desselben verneint haben. Eine andere Ursache ist der Umstand, dass in vielen Fällen der Kern überhaupt nicht in Meiosis eintritt. Dann degeneriert meistens die Zelle allmählich — oft spät — ohne dass der Kern seine Natur geändert hat. Nur in sehr wenigen Fällen dürfte eine somatische Teilung durchgeführt werden. STRASBURGER beobachtete Fälle, mit dem E.M.Z.-Kern in Prophase, wo die Chromosomen von somatischem Aussehen waren. Die

Abbildung, die er gibt, erscheint mir jedoch nicht völlig überzeugend. Die nicht absolut sicheren Fälle, die ich beobachtet habe, wo aus der E.M.Z. ohne vorhergehende Teilung ein Embryosack entsteht, sind vielleicht durch solche somatische Teilungen bedingt — *Antennaria*-Typ. Bei *Chondrilla juncea*, die gleich den apomiktischen *Wikstroemia*-Typen in der Regel dem *Taraxacum*-Schema folgt, hat ROSENBERG (1912) nicht selten »abweichende Fälle» beobachtet. Dort waren die Chromosomen gestreckt, von somatischem Aussehen — ROSENBERGs Abbildung nach zu urteilen. Die Teilung war indessen so spät eingetreten, dass

Entwicklungsstufe	Anzahl Fälle mit								
	E.M.Z. vital, ihr Kern in Ruhe	E.M.Z. degeneriert, ihr Kern in Ruhe	E.M.Z. vital, ihr Kern in Synapsis	E.M.Z. degeneriert, ihr Kern in Synapsis oder auf andere Weise meiotischen Charakter aufweisend	Zellentetraden	Zellendyaden	1-kerniger Embryosack gebildet aus Dyadenzelle	1-kerniger Embryosack gebildet aus ungeteilter E. M. Z.	E. M. Z.-Derivat degeneriert, nähere Angaben können nicht geliefert werden
I	27								
II	20	1	8			11			
III.....................	7	3	2	2	2	10	21	2+3?	2

Vakuolen bereits in der E.M.Z. gebildet waren. Die letztere ist demnach nun als ein junger Embryosack anzusehen. Vermutlich war hier das Resultat Entwicklung nach dem *Antennaria*-Typ, ein Fall analog den möglicherweise bei den apomiktischen *Wikstroemia*-Typen bisweilen vorkommenden.

Über das Aussehen der E.M.Z. oder ihrer Derivate ist die obenstehende Tabelle zusammengestellt worden. Mit I—III werden verschiedene Stufen der Entwicklung bezeichnet. Während I haben die Integumente noch nicht die Nucellusspitze erreicht, die P.M.Z. befinden sich in Ruhe oder in Prophase. Während II haben die Integumente die Nucellusspitze erreicht, die Mikrosporogenese ist im Gange. III ist ein etwas späteres Stadium, die Mikrosporentetraden stehen im Begriff

sich aufzulösen oder haben sich soeben aufgelöst. Die sexuellen *Wik-stroemia*-Arten zeigten Synapsis in der E.M.Z. schon in dem Stadium, das hier mit 1 bezeichnet worden ist.

Die erste Teilung der E.M.Z. trifft also bei der apomiktischen viridi-flora später ein als bei den amphimiktischen Wikstroemia-Arten. Dies geht auch ohne weiteres aus einem Vergleich von Fig. 10 und 11 mit Fig. 6—8 hervor. Bei den letzteren hat in dem Stadium, das dem Stadium II in der Tabelle entspricht, bereits Teilung II stattgefunden, und in dem Stadium, das dem Stadium III entspricht, kommt Vakuole vor und ist demnach die Embryosackbildung eingeleitet worden.

Der reife Embryosack hat dasselbe Aussehen wie bei den sexuellen *Wikstroemia*-Arten. Eine relativ frühe Degeneration der einen Synergide ist nicht beobachtet worden, was darauf deutet, dass ein Pollenschlauch nicht eingedrungen ist. Der Embryo wird aus der Eizelle gebildet. Nucellarembryonie ist nicht beobachtet worden. STRASBURGERS Ablehnung des einzigen von WINKLER vermeintlich angetroffenen Falles von Nucellarembryonie halte ich für wohlmotiviert.

DISKUSSION.

Die hawaiischen *Wikstroemia*-Arten, die ich studiert habe, hatten alle sicher oder wahrscheinlich die Chromosomenzahl n = 9, eine Zahl, die schon für mehrere andere Thymeleaceen bekannt ist (siehe unten). Die Zahl bei *W. indica viridiflora* war dagegen 2n = 27. Diese Zahl liegt sehr nahe der Zahl, nämlich 26, die WINKLER und STRASBURGER als die reduzierte Zahl angaben. Die Verschiedenheit meiner und der früheren Resultate zu erklären, ist ebenso leicht, wie es zu verstehen ist, dass man zu der Zeit, als die agamogonischen Pflanzen zytologisch noch nicht so genau wie jetzt studiert worden waren, den fraglichen Fehler kaum hat vermeiden können. Die Diakinesekerne, in denen die genannten Forscher die Chromosomenzahl bestimmten, waren ganz asyndetisch. Die Chromosomenkörper wurden als Gemini betrachtet, waren aber in Wirklichkeit Univalente. Die Metaphasen in der P.M.Z., die gezählt wurden, waren eben solche, bei denen die Chromosomen »leicht somatisierte» Form hatten und alle Univalente sich genau in eine Äquatorialebene einfügten. Dies geht deutlich aus WINKLERS Abbildungen 15—16 und aus STRASBURGERS Abb. 8 und 10 auf Taf. I hervor. Dass gerade diese Metaphasen gezählt wurden, erklärt sich daraus, dass in den übrigen Fällen strikte Äquatorialplatten nicht gebildet wurden. Dass STRASBURGER später eine Zahl, die um 26 herum pendelte,

sowohl in E.M.Z. wie in Wurzelspitzen fand, was ihn sehr überraschte und zu weitläufigen theoretischen Erwägungen veranlasste, ist nun leicht zu verstehen. Es ist somit vollkommen sicher, dass die Zahl 2n = 27 sowohl dem WINKLERschen wie dem STRASBURGERschen Material eigen gewesen ist.

Die Apomikten unterscheiden sich in der Regel von ihren nächsten normalen Verwandten durch eine grössere Anzahl Genome (vgl. z. B. die Tabellen bei ROSENBERG, 1930 und GUSTAFSSON, 1935 b). *Wikstroemia viridiflora* verhält sich in gleicher Weise, was aus der nachstehenden Tabelle hervorgeht, die die Chromosomenzahlen in der Familie angibt [die Zahlen stammen aus TISCHLERS Tabellen (dort angeführte Literatur) und der vorliegenden Abhandlung]. Nur die 2n-Zahl wird angeführt:

Wikstroemia		*Daphne*		*Gnidia*		*Edgeworthia*	
viridiflora	27	*yezonensis*	18	*carinata*	18	*papyrifera*	36
canescens	18	*alpina*	18				
2737	18	*Mezereum*	18				
2726	18	*Pseudomezereum*	18				
		Kiusiuana	18				
		odora	27?[1]				

Oben ist bemerkt worden, dass allem Anschein nach auch die nicht in der Tabelle angeführten, von mir studierten *Wikstroemia*-Arten diploid sind. Von 7 bekannten *Wikstroemia*-Arten wären demnach 6 diploid und amphimiktisch, 1 triploid und apomiktisch. Innerhalb der nahestehenden Gattungen finden sich offenbar vereinzelte polyploide. Dass die vermutlich triploide *Daphne odora* nicht apomiktisch ist, geht daraus hervor, dass sie regelmässig Makrosporentetraden bildet (OSAWA, 1913). Sie ist stark steril.

STRASBURGER untersuchte den Pollen bei Vertretern der polymorphen Art *W. indica* in den Herbarien BRANDIS' und des Berliner Museums (STRASBURGER, 1909, S. 85 ff.). Zwei Drittel der Exemplare hatten guten Pollen. Diese Individuen stammten aus Sydney, Celebes, Neuguinea, anderen Südseeinseln, Australien und China. Schlechten Pollen hatten dagegen Exemplare von Java, der Bonin-Insel, Srirampur bei

[1] Die Zahl der Annahme nach 27 (vgl. HOLMGREN, 1919). OSAWA bestimmte sie (1913) zu n = 14, später ist sie zu n = 15 und zu 2n = 28 bestimmt worden (vgl. TISCHLERS Tabellen).

Calcutta (derselbe Klon [?] wie der, den WINKLER, STRASBURGER und ich studiert haben). Ähnlichen Untersuchungen habe ich die *Wikstroemia*-Arten im Herbarium des Naturhistorischen Reichsmuseums in Stockholm unterzogen. Normalen Pollen hatten Exemplare von Cobatche und von Nouméa, beide Orte in Neukaledonien, und eines von nicht angegebenem Fundort. Pollen von demselben Aussehen wie bei dem in Buitenzorg vorkommenden Klon hatten ein Exemplar von unbekanntem Lokal und eines von Formosa. Das letztere trug eben die Bezeichnung *viridiflora*. STRASBURGER untersuchte auf dieselbe Weise andere Arten der Gattung, alle hatten normalen Pollen. Von den im Reichsmuseum vertretenen Arten hatten regelmässigen Pollen die mit folgenden Bezeichnungen versehenen: *acuminata, buxifolia, canescens, Caumii, Chamaedaphne, dolichante, furcata, Gampi, haleakabensis* (doch vereinzelte Zwergpollenkörner), *japonica, lichiangensis, nutans, oahensis, ovata, pulcherrima, rotundifolia, stenophylla* und *trichotoma*. Pollen vom »*viridiflora*-Typ» hatte nur *stenantha* von Kwantung. Mehrere der aufgezählten Arten können als richtig bestimmt angesehen werden, mehrere Bestimmungen rühren nämlich von SKOTTSBERG oder von MERRILL her.

Aus diesen Pollenuntersuchungen kann der Schluss gezogen werden, den STRASBURGER (1909, S. 87) zog: von der Art *Wikstroemia indica* gibt es sowohl apomiktische als sexuelle Formen. Dass diese sexuellen Formen nicht triploid sein können, geht daraus hervor, dass sie normalen Pollen haben. Alles spricht demnach dafür, dass die apomiktische *Wikstroemia indica* ein intraspezifischer polyploider Typ ist.

Die Pollenuntersuchungen sprechen dafür, dass nur wenige Arten innerhalb der Gattung durch die Erscheinung gekennzeichnet sind, wie sie bei *Wikstroemia indica* vorkommt.

Die Teilung der Pollenmutterzellen kann bei der studierten *Wikstroemia viridiflora* auf verschiedene Weise geschehen. Man kann eine zusammenhängende Serie von Meiosis mit starker Asyndese bis zu reiner Mitose hin beobachten. Die Beibehaltung der unreduzierten Chromosomenzahl (abgesehen wird hierbei von Komplikationen, erzeugt durch Hemiunivalentenbildung) kann auf dreierlei verschiedene Weise geschehen: 1) durch semiheterotypische Teilung, begleitet von Restitutionskernbildung, 2) durch pseudohomotypische Teilung und 3) durch somatische Teilung. Dass die Chromosomenzahl bei der Bildung des Makrogametophyten wenigstens in der überwiegenden Anzahl

Fälle unreduziert bleiben muss, geht daraus hervor, dass die Pflanze agamospermisch ist, sowie daraus, dass der Embryo sich aus der Eizelle entwickelt. Aus meiner Untersuchung ergibt sich, dass während der Teilung I vorhandene Univalente das Auftreten von Nachzüglern ver-ursachen können. Die Voraussetzung für die Bildung von Restitutions-kernen ist demnach vorhanden. Pseudohomotypische Teilung ist offenbar hier ein anderes Verfahren zur Bildung unreduzierter Embryo-säcke. Dass diese beiden Entwicklungsweisen bei ein und demselben Individuum wirksam sein können, ist von GUSTAFSSON (1935 b) für einige Agamospermen nachgewiesen oder wahrscheinlich gemacht wor-den. Bei anderen, z. B. *Hieracium*, können offenbar sowohl pseudo-homotypische Teilung als auch somatisierte Teilung vorkommen (vgl. GUSTAFSSON, 1935 b; BERGMAN, 1935 b; GENTSCHEFF, 1937). Kann auch die somatische Teilung bei *Wikstroemia* wirksam sein? STRASBURGERS Resultate deuten darauf hin und ebenso mein Nachweis einer evtl. direkten Bildung des Embryosacks aus der E.M.Z., da ja Anlass vor-liegt, zu vermuten, dass das Vorhandensein des *Antennaria*-Schemas ein Kriterium dafür bildet, dass die Teilung »stark somatisiert» gewesen ist, d. h. wenn die Gametophytenbildung einem Entwicklungsmodus homo-log dem Normalschema gefolgt ist. Dass dies hier der Fall ist, kann als sicher angesehen werden, da die übrigen *Wikstroemia*-Arten dem Normalschema folgen. Ich gehe auf die Frage an anderer Stelle des näheren ein (FAGERLIND, 1940).

Es ist also wahrscheinlich, dass dieselben drei Methoden, die zur Bildung unreduzierter Mikrosporen zur Verwendung kommen, auch zur Bildung unreduzierter Embryosäcke verwendet werden. In gewis-sen Fällen konnte die E.M.Z. auch vier Sporen bilden; es zeigt dies, dass noch eine weitere Variation in der ersten Teilung der E.M.Z. vor-kommen kann, eine, die auch bei der Mikrosporenbildung vorhan-den war.

Die apomiktischen Pflanzen, bei denen die Reproduktion über einen Gametophyten geschieht, sind wenigstens in der Mehrzahl der Fälle dadurch gekennzeichnet, dass die Teilung der P.M.Z. und der E.M.Z. sehr verschieden rücksichtlich des Grades der Syndese und des grösseren oder geringeren Grades von meiotischem bzw. mitotischem Charakter verläuft (vgl. beispielsweise GUSTAFSSONs Übersicht 1938). Bei *Wikstroemia indica* (*viridiflora*) kommen indessen, allem nach zu urteilen, genau dieselben Teilungstypen in der P.M.Z. wie in der E.M.Z. vor. Verschiedenheiten in der Frequenz des Vorkommens der einzelnen Typen in den beiden Fällen sind aber deutlich vorhanden. Die Bildung

unreduzierter Derivate ist bei den P.M.Z.-Teilungen ungewöhnlich, bei
den E.M.Z.-Teilungen gewöhnlich. Vermutlich ist der bei vielen Apo-
mikten vorhandene Unterschied zwischen den P.M.Z.- und den E.M.Z.-
Teilungen eben durch einen solchen Frequenzunterschied bedingt. Alle
Arten von Teilungen — verschiedene Grade von Somatisierung —
sind vielleicht immer vorhanden. Hierfür sprechen die Resultate der
Hieracium- und *Antennaria*-Forschung der letzten Jahre (STEBBINS,
1932; BERGMAN, 1935 b—c; GUSTAFSSON, 1935 b und später; GENT-
SCHEFF, 1937). In extremen Fällen sind offenbar gewisse Teilungs-
typen in der P.M.Z. bzw. E.M.Z. so selten geworden, dass sie praktisch
genommen der Beobachtung sich entziehen.

WINGE (1917) und ERNST (1918) stellten die Hypothese auf, dass
Apomixis durch vorhergehende Artbastardierung bedingt wäre. Die
Hypothese wurde lange durch den Nachweis davon gestützt, dass die
Agamospermie oft mit starker Asyndese während der Meiosis verbun-
den war. Später ist nachgewiesen worden, dass Asyndese bei reinen
Arten vorkommen kann. Sie ist da genbedingt (BEADLE, 1930 u. a.).
Mehrere Arten, bei denen agamospermische Formen vorkommen, ent-
halten auch sexuelle diploide Formen. Die Agamospermen müssen da
als intraspezifische, nicht als interspezifische Polyploide betrachtet wer-
den. Der Anschauung, dass die Agamospermie oder wenigstens die
Bildung unreduzierter Gametophyten durch spezifische Gene verur-
sacht sein kann oder verursacht ist, haben sich mehr oder minder ent-
schieden folgende Autoren angeschlossen: ANDERSSON-KOTTÖ, BERGMAN,
GUSTAFSSON, HOLMGREN und ROSENBERG. BERGMAN bemerkt jedoch,
dass, da die Syndeseverhältnisse deutlich auf hybridogene Chromoso-
mengarnitur — das *Drosera*-Schema — hindeuten, doch Bastarde vor-
liegen müssen. Er weist unter anderem auf die *Canina*-Rosen hin.
Hier können die Syndeseverhältnisse geschrieben werden: $7_{II} + 14_I$,
$7_{II} + 21_I$, oder $7_{II} + 28_I$ (TÄCKHOLM, 1922).

Die apomiktische Natur der *Canina*-Rosen ist jedoch nicht nach-
gewiesen. BLACKBURN und HARRISON (1921) wollen die eigentüm-
lichen Verhältnisse bei diesen Pflanzen durch die Annahme erklären,
dass eine Art struktureller Hybridität vorliegt. Diese Ansicht erscheint
auch DARLINGTON (1937) plausibel. Für sie spricht auch, dass, wie
verschiedene Autoren nachgewiesen haben, verhältnismässig gewöhn-
lich 7-chromosomiger Pollen vorliegt. Frühere Kastrationsversuche,
deren Resultate auf apomiktische Vermehrungsweise deuten, haben

allen Wert verloren, seitdem GUSTAFSSON (1931 a—b) gezeigt hat, dass
ein positives Resultat ausbleibt, wenn die Kastrationen sorgfältig aus-
geführt werden. GUSTAFSSON (1937) glaubt jedoch das Vorliegen von
Apomixis (Pseudogamie) durch Bestäubungsversuche mit artfremdem
Pollen beweisen zu können. Er gibt an, dass bei Bestäubung von
Canina-Rosen mit Pollen von *rugosa* und *rubrifolia* die Nachkommen-
schaft einheitlich ausfällt und nicht Charaktere zeigt, die an die des
Pollenlieferanten erinnern. Einen Beweis bilden jedoch diese Resultate
nicht. Da, wenn Befruchtung erfolgt ist, von den 5 Genomen der Nach-
kommenschaft nur eines von dem Pollenlieferanten stammt und die
übrigen vier von der Mutterpflanze, ist es nicht ausgeschlossen, dass die
Eigenschaften des ersteren bei der Nachkommenschaft nicht in Er-
scheinung treten. Um einen schlüssigen Beweis zu erlangen, ist es not-
wendig, die Kreuzungsversuche zu wiederholen, aber unter Verwendung
polyploider Rosen als Pollenlieferanten, wo dann schon die Chromo-
somenzahl bei der eventuellen Nachkommenschaft beweiskräftige Be-
deutung hat. Derartige Kreuzungen unter Verwendung von Pollen von
diploiden, tetraploiden, hexaploiden und oktoploiden Rosen habe ich in
dieser Saison ausgeführt. Ich bin auch an eine zytologische und em-
bryologische Untersuchung der *Canina*-Rosen herangegangen und hoffe
dadurch zu einer Lösung des *Canina*-Problems gelangen zu können.

Sichere Beweise dafür, dass die Apomixis mit Bastardierung in
Zusammenhang steht, liegen also nunmehr kaum vor. Vielmehr kann
man jetzt die polyploiden Agamospermen als intraspezifisch polyploid
betrachten. Wodurch ist dann die Asyndese bedingt? Eine Antwort
glaube ich weiter unten geben zu können.

Beim Studium vor allem von *Galium Mollugo*, aber auch von an-
deren Galieae-Vertretern wies ich nach (FAGERLIND, 1937), dass Tei-
lungen vorkommen, die als ein Mittelding zwischen Meiosis und Mitose
bezeichnet werden können. Bei den studierten Arten fand sich ein viel-
zelliges Makroarchespor, dessen Grenze gegen das somatische Gewebe
sehr diffus war, indem an seiner Peripherie zahlreiche Zellen vorkamen,
die, dem Aussehen nach zu urteilen, weder als somatische Zellen noch
als Archesporzellen bezeichnet werden konnten. Sie bildeten vielmehr
eine Übergangsserie zwischen diesen beiden Zellarten. Die Teilungen
im Zentrum des Archespors waren Meiosen mit normalen Syndesever-
hältnissen. Nach der Peripherie des Archespors hin wurde die Syndese
mit dem Abstand vom Zentrum schwächer und schwächer. Die Chro-
mosomenform stimmte dagegen stets mit der Chromosomenform wäh-
rend Teilung I einer gewöhnlichen Meiosis überein. An den äussersten

Grenzen des Archespors und ausserhalb desselben kam ein ähnlicher Teilungstyp vor, aber mit noch stärkerer Asyndese, recht oft mit totaler. Die mehr oder weniger asyndetischen Teilungen wurden als semihetero-typische durchgeführt. Ein Vorkommen von pseudohomotypischer Teilung war jedenfalls möglich. Sie ist jedoch nicht mit voller Sicher-heit nachgewiesen worden. Während aller dieser Teilungen lag trotz der Asyndese volle Chromosomenhomologie vor. Die Asyndese ist dem-nach hier nicht durch den Mangel einer solchen Homologie bedingt. Auch kann sie nicht genbedingt sein, da im Zentrum des Archespors und während der Mikrosporogenese die Syndese normal war (abgesehen wird hier von den abnormen Fällen, die ich auch nachgewiesen habe). Sie muss also darauf beruhen, dass die Teilung nicht eine heterotypische war. Die Teilung muss als ein Übergang zwischen heterotypischer Teilung und Mitose — als eine »somatisierte Meiosis» oder als eine »meiotisierte Mitosis» — betrachtet werden.

Fälle, die an die »somatisierten Meiosen» bei den Galieae-Vertretern erinnern, sind auch anderwärts angetroffen worden. Ich denke an die Teilungen, die ROSENBERG (1909) und ich selbst (FAGERLIND, 1937) als Abnormitäten in den Tapetumzellen oder anderen benachbarten somati-schen Zellen der Staubbeutel nachgewiesen haben. Diese Teilungen waren durch die meiotisch kontrahierte Form der Chromosomen, aber fehlende Syndese ausgezeichnet. Die Teilungen erfolgten semihetero-typisch.

»Somatisierung» der Meiosis kann also eine Teilung mitführen, bei der die Chromosomen mit denen der Meiosis betreffs der Form überein-stimmen, bei der Syndese vorkommen kann, diese aber mehr oder minder abgeschwächt ist. Totale Asyndese kann oft die Folge sein. Die »somatisierte Meiosis» kann dem semiheterotypischen oder dem pseudohomotypischen Schema folgen. Solche Teilungen bei den Aga-mogonen als durch vorhergehende Bastardierung bedingt anzusehen, ist man also kaum mehr berechtigt. Diese Teilungen können hier ebensogut durch genbedingte »Somatisierung» der Meiosis verursacht sein. Da wir sicher wissen, dass mehrere Agamospermen nicht Bastarde sind, und ferner dass mehrere Teilungen aufweisen, die durch starke Somatisie-rung ausgezeichnet sind, halte ich den folgenden Schlussatz für be-rechtigt:

Die Asyndese bei vielen Agamogonen ist sehr wahrscheinlich durch die »Somatisierung» der Meiosis bedingt. Ist diese stark, so werden Teilungen erhalten, bei denen die Chromosomen somatische Gestalt haben; ist sie schwach, so behalten die Chromosomen ihre meiotische

*Gestalt, aber es tritt Asyndese ein. Ist die »Somatisierung» sehr schwach,
so wird die Teilung sehr ähnlich einer Meiosis (semiheterotypische
Teilung); ist sie etwas stärker, so wird die Asyndese total. Pseudohomo-
typische Teilung kann dann eintreten.*

— — —

Bei den Amphimikten geschehen in der Regel (betreffs weniger
Ausnahmen siehe unten), wenn die Entwicklung von E.M.Z. zu Sporen-
bildung und ferner zu Gametophytenbildung und -entwicklung geht, der
Reihe nach folgende Veränderungen:

I	II	III	IV
heterotypische	homotypische Teilung	Wachstum und	somatische Tei-
Teilung	→ (=›Äquationsteilung», →	Vakuolenbil-	→ lung Nr. 1	→ usw.
	durchgeführt mit kon-	dung	
	trahierter Chromoso-		
	menform)		

Bei Agamogonen mit Semiaposporie (eine oder mehrere Arten der
Gattungen *Taraxacum, Wikstroemia, Erigeron* u. a. — vgl. Tabelle 2
bei FAGERLIND, 1940 und dort angeführte Literatur) wird die erste
Teilung der E.M.Z. als pseudohomotypisch durchgeführt. Bei diesen
geschehen der Reihe nach folgende Veränderungen:

I	II	I
pseudohomotypische Teilung (= Äqua-	Wachstum und	somatische Tei-
tionsteilung, gekennzeichnet durch kon- →	Vakuolenbildung →	lung Nr. 1	→ usw.
trahierte Chromosomen)		

Bei Agamogonen mit Aposporie (eine oder mehrere Arten der Gat-
tungen *Antennaria, Eupatorium, Hieracium* u. a. — vgl. Tabelle 2 bei
FAGERLIND, 1940 und dort angeführte Literatur) wird die erste Teilung
der E.M.Z. als rein somatisch durchgeführt. Bei diesen geschehen der
Reihe nach folgende Veränderungen:

I	II
Wachstum und Vakuolenbildung →	somatische Teilung Nr 1 → usw.

In dem ersten der hier für die Agamogonen angeführten Fälle ist
also die Entwicklung, verglichen mit den Verhältnissen bei den Amphi-
mikten, um einen Teilungsschritt (heterotypische Teilung eliminiert),
in dem anderen um zwei Teilungsschritte (heterotypische und homo-
typische Teilung eliminiert) verkürzt worden. Es ist auffallend, dass
in den drei Fällen die vorkommenden Entwicklungsphasen genau die-
selbe Reihenfolge innehalten. In analoger Weise, wie die Spore sich in
den normalen Fällen entwickelt, entwickelt sich also die Dyadenzelle

(oder das Produkt, der Dyadenkern), wenn Semiaposporie vorliegt, und die Mutterzelle selbst, wenn Aposporie vorliegt. Wenn Semiaposporie vorliegt, hat sich offenbar die Dyadenzelle in einen jungen Embryosack umgewandelt; wenn Aposporie vorliegt, wandelt sich die Mutterzelle direkt in einen jungen Embryosack um. Liegt Aposporie vor, so hat sich also der Embryosackmutterzellkern direkt in den Embryosackkern umgewandelt. Wenn Semiaposporie vorliegt, hat sich offenbar der E.M.Z.-Kern so umgewandelt, dass er mit dem Dyadenkern bei normaler Entwicklung übereinstimmt. In Anbetracht der analogen Entwicklung kann man kaum einen anderen Schluss ziehen. Ist er richtig, so muss die pseudohomotypische Teilung als ihrer Natur nach mit der homotypischen gleichartig betrachtet werden. Oben habe ich Tatsachen angeführt, die dafür sprechen, dass die pseudohomotypische Teilung ihrer Natur nach ein Mittelding zwischen somatischer Teilung und heterotypischer Teilung darstellt. Ist das der Fall, so ist damit auch der Schluss gegeben, dass die homotypische Teilung ihrer Natur nach eine solche intermediäre Erscheinung ist.

Für *Wikstroemia* ist oben gezeigt worden, dass bei Klonen, wo die Gametophytenbildung in der Regel durch Semiaposporie zu geschehen scheint, die Entwicklung des E.M.Z.-Kerns eine Verzögerung erfährt, verglichen mit der bei nahestehenden Amphimikten. Die Verzögerung ist derart, dass die Vakuolenbildung in der Dyadenzelle, d. h. die Gametophytenbildung, ungefähr gleichzeitig damit geschieht, dass diese Erscheinung bei den Amphimikten stattfindet. Bei Apomikten mit generativer Aposporie scheint die Verzögerung der ersten Teilung der E.M.Z. noch grösser zu sein als bei *Wikstroemia* (vgl. BERGMAN, 1935 b—c; STEBBINS, 1932; AFZELIUS, 1936; GUSTAFSSON, 1935—39). Im letzteren Falle war, verglichen mit den Verhältnissen bei den Amphimikten, eine Teilung »eliminiert» worden, im ersteren zwei. In beiden Fällen dürften also Vakuolisierung (die Gametophytenbildung) und die erste Teilung des Gametophyten ungefähr zu gleicher Zeit wie bei nahestehenden Amphimikten eintreffen. Eine Verzögerung ist meines Wissens zuvor nicht beobachtet worden, wenn Semiaposporie vorliegt. Es beruht dies vermutlich darauf, dass die Verzögerung in diesem Falle nicht so ausgesprochen ist, wie wenn generative Aposporie vorhanden ist.

Auf Grund der eben angeführten Verhältnisse hat GUSTAFSSON (1935, 1938, 1939) eine Hypothese über die Interrelation und Physiologie der Kernteilungen aufgestellt. Leider ist es recht schwer, überall zu verstehen, was er meint. Er scheint geltend machen zu wollen, dass die Meiosis sich von der Mitose durch andere Wassergehalts- und

Viskositätsverhältnisse unterscheidet, dass die Meiosis sich gradweise (?)
in Mitose umwandelt durch Veränderung des Grades von »hydration»,
»sap-intake» und Viskosität des Kerns und der Zelle, Dinge, von denen
wir — und auch GUSTAFSSON — absolut nichts wissen. Entscheidend für
die Beurteilung hier scheinen die Volumveränderungen der E.M.Z. und
ihres Kerns und der Vakuolisierungsgrad des ersteren zu sein. Es ist
möglich, dass ich GUSTAFSSONs Ausführungen missverstehe und daher
seine Hypothese — die indessen eher als eine erwiesene Tatsache dar-
gestellt wird — nicht ihrem vollen Werte nach würdigen kann. In seiner
letzten Arbeit geht jedoch GUSTAFSSON (1939) noch weiter. Hier will er
Embryosackentwicklung nach dem *Scilla*- und *Lilium*-Typ (= tetra-
sporische Embryosacktypen?) als Zwischenstadien zwischen Entwick-
lung nach dem Normaltyp und nach dem *Taraxacum*- und *Antennaria*-
Typ einschalten. Bei Entwicklung nach dem Normaltyp machten sich,
meint er, »vacuolisation forces» erst geltend, nachdem »meiotic sub-
stances» Meiosis hervorgerufen hätten und die Entwicklung bis zur Bil-
dung von Zellentetraden fortgeschritten sei. Wenn »vacuolisation forces»
sich schon nach der Bildung der Zellendyade geltend machten, sei das
Resultat *Scilla*-Typ; wenn sie sich gleich nach dem Beginn der meioti-
schen Prophase geltend machten, sei das Resultat *Lilium*-Typ; wenn sie
sich schon vorher geltend machten, sei das Resultat, dass die Meiosis
in E.M.Z. sich in eine pseudohomotypische oder somatisierte Teilung
umwandle. Leider stimmen jedoch die Tatsachen durchaus nicht mit
den Behauptungen überein, auf die seine Hypothesen sich stützen. In
der Regel treten Vakuolen erst auf, nachdem vier Sporenkerne gebildet
worden sind, gleichgültig ob die Entwicklung einem der mono-, bi- oder
tetrasporischen Schemata gefolgt ist. Hier die ganze neuere embryologi-
sche Literatur heranzuziehen, um GUSTAFSSON zu widerlegen, der selbst
nicht durch Literaturzitate zeigt, woher er seine Angaben genommen
hat (es sieht aus, als stammten sie lediglich aus Referaten von MODI-
LEWSKIs und RUTGERs veralteten Übersichten), erscheint mir unnötig.
Von der Regel gibt es indessen einige wenige Ausnahmen (*Adoxa*,
Helosis, *Limnanthes*, *Tulipa*, um ein paar Beispiele zu nennen). Diese
stehen nicht in irgendwelchem nachweisbaren Zusammenhang mit dem
Entwicklungstyp, wenn auch die Ausnahmen vielleicht nicht unter den
monosporischen Typen vorkommen. Bei einigen der Ausnahmen wer-
den Vakuolen sehr früh angelegt. Die Meiosis ist demungeachtet nor-
mal. Frühe Vakuolenbildung ist demnach nicht Ursache oder Kriterium
einer »somatisierten» Teilung.

In welchem gegenseitigen Zusammenhang die bei Aposporie und

Semiapasporie nachgewiesene Entwicklungsverspätung, die »Somatisie-
rung» der E.M.Z.-Teilung und Vakuolenbildung zueinander stehen, lässt
sich nicht entscheiden. Gegenwärtig auf Grund morphologischer Beob-
achtungen weitgehendere Schlüsse betreffs der Physiologie der Kern-
teilungen ziehen zu wollen, kann ohne weiteres als vollkommen zweck-
los bezeichnet werden.

Vor dem Abschluss dieser Arbeit ist es mir eine angenehme Pflicht,
allen denen, die mir bei der Beschaffung des Materials behilflich gewe-
sen sind oder in anderer Weise meine Arbeit unterstützt haben, meinen
verbindlichen Dank auszusprechen. Unter diesen seien besonders er-
wähnt Herr Dr. TH. VAN DEN HONERT (Buitenzorg), Herr Professor
O. ROSENBERG (Stockholm), Herr Professor C. SKOTTSBERG (Göteborg)
und Herr Dr. D. F. VAN SLOOTEN (Buitenzorg).

ZUSAMMENFASSUNG.

1. Die Grundzahl bei der Gattung *Wikstroemia* ist 9.

2. Ein studierter agamogonischer Klon von *Wikstroemia viridiflora*
(*indica*) ist intraspezifisch triploid.

3. Triploidie und Agamogonie sind selten innerhalb der Gattung
Wikstroemia.

4. Die meisten *Wikstroemia*-Arten haben normale Pollenbildung
und folgen in ihrer Embryosackbildung dem Normaltyp.

5. Bei dem studierten agamogonischen *W. viridiflora*-Klon sind
während der Mikrosporogenese sehr verschiedene Teilungstypen vorhan-
den. Eine stetige Reihe von relativ stark syndetischer Meiosis über
stark oder total asyndetische Meiosis und pseudohomotypische Teilung
zu rein somatischer Teilung hin ist nachgewiesen worden.

6. Bei dem agamogonischen Klon zeigt die E.M.Z., allem nach zu
urteilen, dieselben Variationen wie die P.M.Z. Am gewöhnlichsten ist
jedoch wohl pseudohomotypische Teilung oder semiheterotypische Tei-
lung kombiniert mit Restitutionskernbildung.

7. Bei dem agamogonischen Klon liegt meistens (?) Semiapo-
sporie vor, die Embryosackbildung folgt hauptsächlich dem *Taraxacum*-
Schema. In vereinzelten Fällen können reduzierte Embryosäcke gebil-
det werden. Dann treten während der Entwicklung Zellentetraden auf.

8. Die Asyndese während der Meiosis bei Agamogonen wird als
durch genbedingte »Somatisierung», nicht durch mangelnde Homologie

verursacht angenommen. Die polyploide, agamogonische Form wird in der Regel für intraspezifisch polyploid erklärt.

9. Es wird wahrscheinlich gemacht, dass die pseudohomotypische und die homotypische Teilung ihrer Natur nach gleichartig sind.

10. Ein gegenseitiger Zusammenhang zwischen Vakuolenbildung und Embryosackbildung gemäss dem Normalschema, den bisporischen, den tetrasporischen Schemata, dem *Taraxacum*- oder dem *Antennaria*-Schema liegt nicht vor.

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POLYPLOIDY IN PARTHENOGENETIC CURCULIONIDAE

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(Preliminary Report)

PARTHENOGENETIC reproduction is very rare in *Coleoptera*, contrary to many other orders of insects (cf. SZÉKESSY, 1937). Most of the parthenogenetically reproducing species of beetles belong to the weevils, *Curculionidae*. Within this group parthenogenesis is comparatively common in the subfamilies *Otiorrhynchinae* and *Brachyderinae*. In other families of *Coleoptera*, however, only a few parthenogenetically reproducing species are at present known. As bisexual as well as parthenogenetic species are found in many genera of the above-mentioned curculionids, a study of their cytology affords an excellent opportunity of comparing the chromosomes in closely related bisexual and parthenogenetic species of animals. In the following pages a brief summary of some results of my studies concerning these matters will be given.

The material used was collected in different parts of Finland. It was fixed in CARNOY's fluid (6 : 3 : 1) and sectioned. The preparations were stained in HEIDENHAIN's iron-hematoxylin or with the fuchsin sulphurous acid of FEULGEN. The drawings were made with the aid of an objective 120, ocular 25 and Abbe's camera lucida (all »Zeiss»). The magnification is about 3000 $\times$.

I. THE BISEXUAL SPECIES.

Since, as far as I am aware, nothing has been published about the cytology of true curculionids, I have also studied some bisexual species; they have as far as possible been selected from the same genera as the parthenogenetic species. The following results were obtained.

Strophosomus capitatus (= *Str. rufipes* v. *capitatus*). — The diploid chromosome number of the species is 22. In the male there are 11 chromosome bodies of different size in the metaphase of the first meiotic division (Fig. 1). The smallest is the unpaired X-chromosome, but ten are bivalents. The X precedes the others in the anaphase.

Thus in the metaphase of the second division there are two kinds of cells with respect to their chromosome complements, *viz.* one with 11 and the other with 10 chromosomes. The males are thus heterogametic and belong to the *XO-type*. I have not been able to obtain preparations of male somatic divisions. In the female the metaphase of the first meiotic division (Fig. 2) shows 11 bivalents. The X-pair is smaller than the other bivalents. The oogenesis is completely normal. The females are thus homogametic. In the metaphases of female somatic divisions 22 long, often V-shaped, chromosomes can be seen (Fig. 3). Both X-chromosomes differ from the autosomes in being of a smaller size.

*Otiorrhynchus arcticus.* — The diploid number of chromosomes
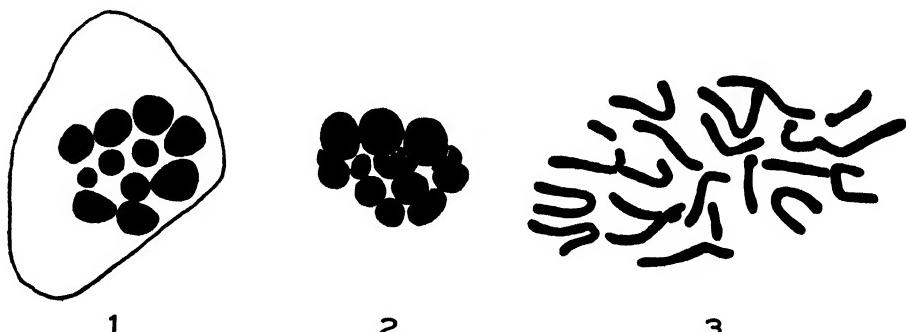
Figs. 1—3. *Strophosomus capitatus.* — Fig. 1. ♂, first metaphase. — Fig. 2. ♀ first metaphase. — Fig. 3. ♀, blastomere mitosis.

is 22. In the metaphase of the spermatogonia (Fig. 4) 22 chromosomes appear, one of which, the Y-chromosome, is considerably smaller than the others. The males of this species are thus of the *XY-type*. The X and Y pair in the first meiotic division, and so there are 11 bivalents of different size in the metaphase plate (Fig. 5). X and Y do not precede the other chromosomes in this species. There are two kinds of metaphase plates of the second division; in some the larger X and in others the smaller Y is present. In the female the metaphase of the first meiotic division also (Fig. 6) shows 11 bivalents. The paired X-chromosomes separate first (cf. the figure). As in the two following species I have not been able to obtain satisfactory preparations of somatic divisions.

Polydrosus pilosus. — The diploid number of chromosomes is 22. The chromosome behaviour in this species is similar to that of *Otiorrhynchus arcticus*. The X- and Y-chromosomes appear paired in the

first meiotic division, there being thus 11 bivalents in the metaphase
plate. In the metaphase plates of the second division either the larger
X or the smaller Y is present. I did not obtain any clear preparations
of the female meiosis, for the chromosomes are so closely packed
together that no exact counts can be made. In another *Polydrosus*
species, *P. undatus*, I have, however, ascertained beyond doubt the
presence of 11 bivalents in the metaphase of the first meiotic division
in the female.

Hylobius abietis. — This weevil, which is more remotely related to
the last mentioned species, also has the diploid number 22. With
regard to the chromosomes the species is similar to *Strophosomus
capitatus*, both belonging to the *XO-type*. In the metaphase of the first
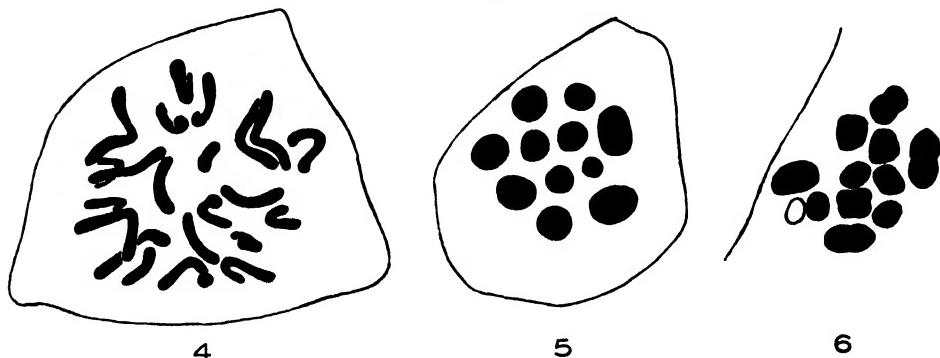
Figs 4—6. *Otiorrhynchus arcticus*. -- Fig. 4. Spermatogonial metaphase; Y in the
middle. — Fig. 5. ♂, first metaphase. — Fig. 6. ♀, first metaphase; the X-chromo-
somes (on the left, bottom) have already separated.

meiotic division in the male there are 11 chromosome bodies of differ-
ent size. The smallest is the unpaired X-chromosome; it clearly precedes
in the first anaphase.

To sum up, it may be said that *all the bisexual species of weevils
studied by me have the same number of chromosomes* (2n = 22). The
males are heterogametic, belonging either to the XO- or the XY-type;
the females are homogametic.

II. THE PARTHENOGENETIC SPECIES.

It is to be expected that the cytology of parthenogenetic weevils
is not devoid of interest from more than one point of view, for it
should be noted that many of the species in question reproduce
parthenogenetically within definite areas only, being bisexual in other

areas. On the other hand, if we examine exclusively parthenogenetic species we shall find that many of them have closely related bisexual species characterized only by slight external differences, having distribution areas different from those of the former and usually south of them. For further information on these problems the reader is referred to papers, for instance, by PENECKE (1922) and SZÉKESSY (1937). It may be pointed out that VANDEL (1932), in discussing the *spanandrie* and parthenogenesis in the weevils of the genus *Trachyphloeus*, expresses the opinion that the parthenogenesis is accompanied by polyploidy.

The chromosome numbers of the nine parthenogenetic species of curculionids studied by me, form an arithmetic series. *They are 22, 33 and 44* or at any rate a number very close to one of these figures. Since the diploid number of all the bisexual species examined is 22, it is obvious that *among the parthenogenetic species there are, in addition to diploid species, also triploid and tetraploid.*

1. THE DIPLOID SPECIES.

Only one of the species examined belongs to this group.

*Polydrosus mollis.* — In the metaphase plates of the mature eggs
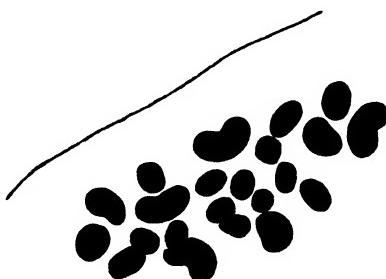
Fig. 7. *Polydrosus mollis.* Metaphase of maturation division, 22 chromosomes.

22 chromosomes are plainly visible (Fig. 7). They are of different size, four being evidently larger than the others. The egg-cell passes through one maturation division only, which is equational. The somatic cells thus retain the normal diploid number, a fact I have verified.

2. THE TRIPLOID SPECIES.

Five of the examined species belong to this group.

Otiorrhynchus ovatus. — The egg-cells of this species also pass

through one maturation division only, which is equational.   There is accordingly no reduction.   The number of chromosomes in the meta-
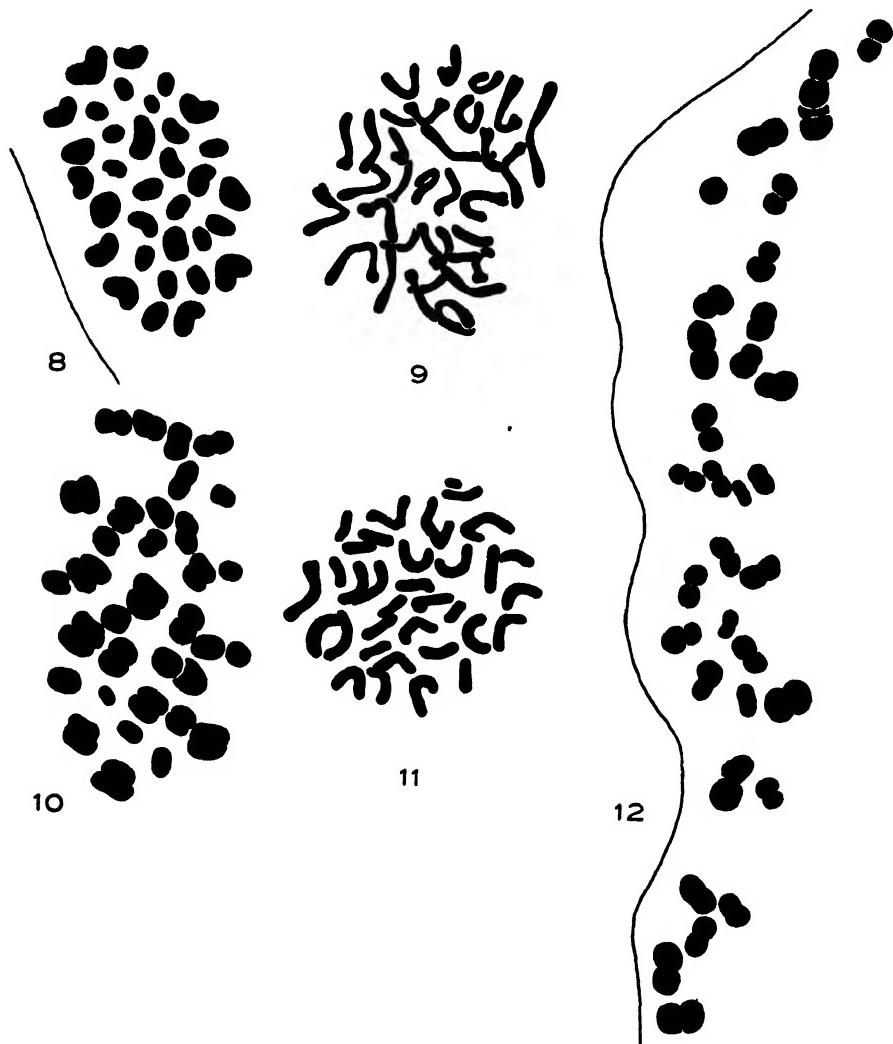
Figs. 8—9. *Otiorrhynchus ovatus.* 33 chromosomes. — Figs. 10—11. *Strophosomus melanogrammus.* 34 chromosomes. — Fig. 12. *Sciaphilus asperatus.* 33 chromosomes. — Figs. 8, 10 and 12 metaphase of maturation division.   Figs. 9 and 11 blastomere mitosis.

phase plates of the maturation divisions varies between 30 and 34.   This variation can be observed even among the eggs of one and the same female.   I do not intend to discuss its causes in this connection.   The

most common chromosome numbers are 32 and 33. The chromosomes
are of varying size, more or less globular or somewhat oval (Fig. 8).
Also the size of the plates varies considerably, the younger plates
probably being larger. The maturation division being equational, the
somatic cells retain the same number of chromosomes as the meta-
phase of the maturation division. I have not been able to ascertain
whether the number of chromosomes of *Otiorrhynchus ovatus* varies
in somatic cells also, because of the difficulties of making exact counts
in the latter. In the somatic metaphase shown in Fig. 9 there are 33
long chromosomes, bent in the middle. They vary in size, but the
different chromosome types are indistinguishable.

Otiorrhynchus ligustici. — The eggs of this species, rare in Fin-
land, are comparatively large and difficult to cut and therefore satis-
factory preparations are not easily obtained. For that reason I have
so far been unable to determine the chromosome number exactly. In
four metaphase plates of the maturation division I found about 33—35
chromosomes. It is noteworthy that in some eggs the chromosomes
are in two different groups, one of which contains the diploid, the other
the haploid number of chromosomes. (Cf. the cytology of *Otiorrhyn-
chus dubius*, below). In the light of all this *Otiorrhynchus ligustici*
must in my opinion be regarded as clearly triploid.

Strophosomus melanogrammus. — The egg-cells of this species
also pass through one maturation division only, which is equational.
The number of chromosomes in the metaphase of the maturation
division varies, as in *Otiorrhynchus ovatus,* being 31—35. The most
common number is 34 (Fig. 10). The size of the chromosomes in one
plate varies considerably; in every plate there is one chromosome
evidently smaller than the others. The equational split is often very
distinct. The maturation division begins while the egg is still in the
ovary, the eggs being at early anaphase when laid. The number of
chromosomes remaining in the egg after the maturation division is the
same as in the metaphase. In this species, too, I was not able to find
out whether the chromosome number varies in somatic divisions. In
the somatic metaphase pictured in Fig. 11 there are 34 chromosomes.
The smallest one plainly differs from the others (top of the figure).

Trachyphloeus bifoveolatus. — I have been unable to determine
the exact number of chromosomes of this species, though I have
examined fifteen sectioned ovaries. At metaphase the chromosomes
are attached to each other, forming long, branched chain-like
structures, the boundaries of the different chromosomes being difficult

to observe.   In one metaphase plate I was able with a rather high degree of accuracy to count 32 chromosomes.   In addition to this I also counted a little more than 30 chromosomes in four more plates.   The triploid number of chromosomes is thus obvious in this species too.

Sciaphilus asperatus. — The chromosome number of this species is 33.   In the metaphase of the maturation division 33 chromosomes of different size are to be seen (Fig. 12); in them the equational split is nearly always visible.   The chromosomes divide while the egg is still in the ovary.   The chromatids begin to separate in the plane of the plate, so that one usually finds plates which seem to have more than 33 chromosomes.   The sister chromatids, however, usually lie so clearly parallel that 33 chromosomes or chromatid pairs can also be ascertained in these cases.   In one egg I found two metaphase plates, one of which had 22, the other 11 chromosomes.   (Cf. the cytology of *Otiorrhynchus dubius.*)   In the somatic cells I was able to ascertain the triploid number of chromosomes.

3. THE TETRAPLOID SPECIES.

Three of the species studied by me belong to this group.

Otiorrhynchus dubius. — The chromosome number of this species is 44.   I have not yet succeeded in ascertaining whether there is in this species a variation of the chromosome number corresponding to that found in certain other parthenogenetic species.   It is interesting to note that in *Otiorrhynchus dubius* there are even in the same ovary several types of egg-cells different with respect to the grouping of the chromosomes in the metaphase of the maturation division.   Four kinds of egg-cells can be distinguished:

a) Egg-cells with all the 44 chromosomes in one group (Fig. 13). The metaphase plates thus show the *tetraploid number.*   The chromosomes vary in size, though the different types of the chromosomes cannot be distinguished.

b) Egg-cells, in which the chromosomes form two different groups. In one plate there is the *triploid number (33)* and in another the *haploid number (11)* of chromosomes (Fig. 14).

c) Egg-cells, the chromosomes of which are in two distinct groups, but with the *diploid (22) number* of chromosomes *in each* (Fig. 15).

d) Egg-cells, in which there are three groups of chromosomes. *One plate* has in this case always the *diploid number (22)* and the *two others the haploid number (11)* of chromosomes.

These different chromosome plates are sometimes quite close to each other, but they may also be comparatively far apart. Their relative position also varies; they may be parallel, but also at right angles to each other. — In some eggs the chromosomes were in irregular groups.

As already described, I found a corresponding phenomenon — although less commonly — in a few other parthenogenetic curculionids, *viz. Otiorrhynchus ligustici, Otiorrhynchus scaber* and *Sciaphilus asperatus.*

I have not so far been able to find out how the maturation division

13   **14**   **15**
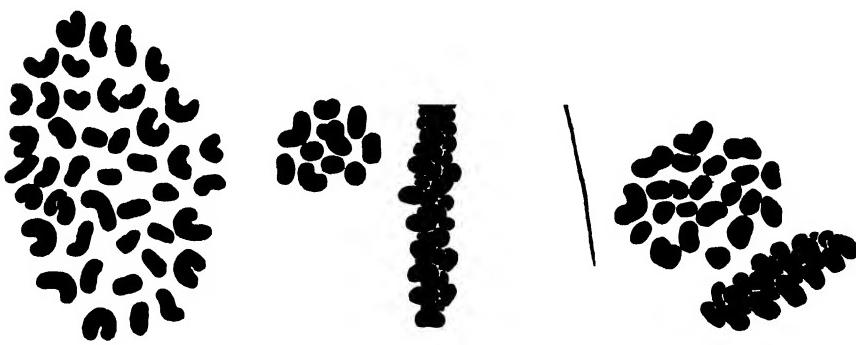
Figs. 13—15. *Otiorrhynchus dubius.* Metaphase of maturation division, 44 chromosomes. — Fig. 13. Tetraploid plate. — Fig. 14. Triploid and haploid plate in the same egg. — Fig. 15. Two diploid plates in the same egg.

and blastomere divisions take place in the eggs of different types in *Otiorrhynchus dubius.*

Otiorrhynchus scaber. — The egg-cells of this species pass through a single maturation division only, which is equational. The chromosome number in the metaphase plates of this division varies, as in *Otiorrhynchus ovatus* and *Strophosomus melanogrammus,* being 42—44. The most common number is 44 (Fig. 16). The chromosomes are relatively large and of varying size. Their shape is globular or somewhat oval. — Sometimes, but very rarely, I found two distinct groups of chromosomes. In these cases there was always found either the diploid number in both plates or the triploid number in one and the haploid in the other plate. (Cf. the cytology of *Otiorrhynchus dubius,* above.) — In the somatic cells of *Otiorrhynchus scaber* I was able to ascertain the tetraploid number of chromosomes.

Barynotus obscurus. — In the mature eggs of this species 44 chromosomes can be clearly seen (Fig. 17). They are very large, larger than in any of the other weevils studied. Their size is variable. In the middle they often show a clear constriction. I did not succeed in obtaining any preparations of the somatic divisions of this species.

In summarising the cytology of the investigated parthenogenetic curculionids the following facts may be pointed out:

Polydrosus mollis is *diploid.*

Otiorrhynchus ovatus, O. ligustici, Strophosomus melanogrammus, Trachyphloeus bifoveolatus and *Sciaphilus asperatus* are *triploid.*

Otiorrhynchus dubius, O. scaber and *Barynotus obscurus* are *tetraploid.*
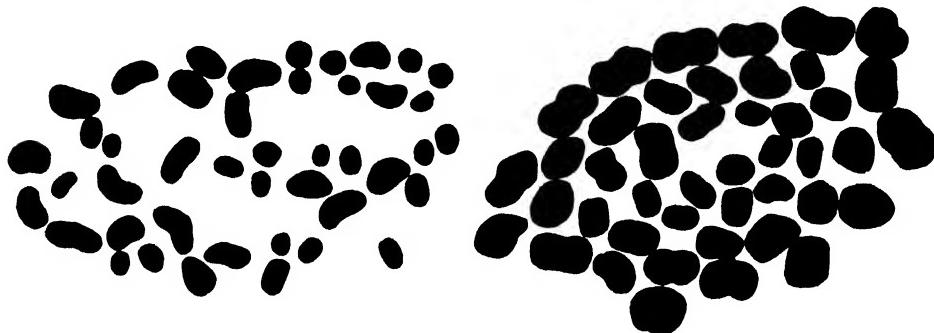

Fig. 16. *Otiorrhynchus scaber.* Metaphase of maturation division, 44 chromosomes. — Fig. 17. *Barynotus obscurus.* Metaphase of maturation division, 44 chromosomes.

The egg-cells pass through one maturation division only, which is equational. It is noteworthy that in some species *the chromosomes at the metaphase of the maturation division may be arranged in two or even three different plates, each plate containing one or more complete sets of chromosomes.*

III. DISCUSSION.

Although a common phenomenon in plants, polyploidy is comparatively rare in the animal kingdom; see, for instance, KAWAGUCHI (1936, p. 129—130) and DOBZHANSKY (1937, p. 219—224). In a great many cases polyploidy appears in connection with parthenogenesis. Well known are the tetraploid and octoploid parthenogenetic races of *Artemia salina* (ARTOM, 1911; GROSS, 1932), the triploid partheno-

genetic race of *Trichoniscus provisorius* (VANDEL, 1928), and the tetra-ploid parthenogenetic races of *Solenobia triquetrella* and *S. lichenella* (SEILER, 1923 and 1939). All these species have in addition a diploid bisexual and some also a diploid parthenogenetic race.

MULLER (1925) assumed that the rarity of polyploidy in animals and its abundant occurrence in plants is due to the fact that most higher plants are monoecious, while animals are dioecious. Since the sex-determination in the latter is generally regulated by a sex-chromo-some mechanism, polyploidy may upset this mechanism and thus give rise to sexually abnormal and sterile forms (cf. also DOBZHANSKY, 1937, p. 219—220). This danger is not present in parthenogenetically reproducing animals, the egg-cells of which have an equational division only, thus also in the parthenogenetic curculionids described in this paper. In these the chromosome sets and, accordingly, the relationship between sex-chromosomes and autosomes naturally remain unchanged.

It is a well known fact that many parthenogenetic species of plants are polyploid, as compared with other closely related bisexual species. WINGE (1917) and ERNST (1918) simultaneously advanced the view that polyploidy and, in connection with it, parthenogenesis have arisen as a consequence of the crossing of species. This assumption has been found to hold good with regard to many plants; see, for instance, ROSENBERG (1930, p. 57—59). ROSENBERG (p. 58) remarks, however: »Man kommt vielleicht den wahren Verhältnissen näher, wenn man die Sache so ausdrückt, dass die Bastardierung nicht die Ursache der Parthenogenesis ist, sondern sie kann sehr wohl ein wich-tiges Moment sein für die Entstehung parthenogenetischer Formen und vielleicht sogar eine Voraussetzung dafür, ohne dass jedoch ein eigent-liches Kausalverhältnis zwischen den beiden Erscheinungen bestände». The tendency to parthenogenetic development is, according to HOLM-GREN (1919, p. 112), inherent in the gametes, but it may not be able to take effect in haploid ones.

When looking for the causes of polyploidy and parthenogenesis in animals, we must no doubt begin with the assumption that these phenomena can be produced in different ways in different forms. SEILER (1923, p. 83—84) is of opinion that as regards the partheno-genetic races of *Solenobia* »der Gedanke einer Artbastardierung als Ursache der Parthenogenese ausgeschlossen ist». He thinks (1923 and 1939) that in *Solenobia* the parthenogenesis originally arose in a di-ploid form (there is a diploid parthenogenetic race of *Solenobia tri-quetrella*) and that the chromosome number subsequently became

doubled by automixis or a similar fusion of two diploid nuclei. Something of this kind has been found by GROSS (1932) in *Artemia*. The polyploid races of *Solenobia* and *Artemia* should thus be autopolyploid. VANDEL (1928 and 1931) is of opinion that the polyploidy and parthenogenesis in *Trichoniscus*, too, have not arisen as a consequence of crossing, but more probably through the fertilisation of a diploid egg. VANDEL (1931, p. 318) says: »On voit donc que jusqu'ici l'idée de l'origine hybride des formes parthénogénétiques constitue, au moins dans le règne animal, une simple hypothèse de travail». HEILBORN (1934, p. 236) also remarks that »the polyploidy hitherto known among animals is autopolyploidy . . .; animal allopolyploidy is as yet unknown».

The question arises as to how the parthenogenesis and polyploidy in the curculionids arose. It seems clear to me that they need not necessarily have arisen simultaneously but that parthenogenesis may also arise in a diploid form (*e. g. Polydrosus mollis*), an opinion held also by SEILER (1923 and 1939) in the case of *Solenobia*. If we consider the behaviour of the chromosomes in the polyploid parthenogenetic curculionids, we find that at least in four out of nine species (most clearly in *Otiorrhynchus dubius*) there appears in the metaphase of the maturation division a kind of gonomery. This seems to indicate that *in these curculionids the polyploidy has arisen as a result of crossing*, that is to say, *these species are allopolyploid*. The fact that in triploid species a complete set of chromosomes and in tetraploid species one or two sets may be separated from the rest of the chromosomes, proves in my opinion, that the sets in question are secondary and extraneous in origin. A new haploid set of chromosomes of this kind apparently does not always form a completely harmonious whole with the original diploid set, which always remains one single plate. Some influence may move it away from the latter, nevertheless keeping its chromosomes together. Evidence in favour of allopolyploidy is also afforded by the fact that the chromosome complement of *Strophosomus melanogrammus* contains one small chromosome different from the others, instead of three, as we should expect if the species was autopolyploid. It seems to me most probable that the parthenogenesis of curculionids originally arose in diploid forms. We may further assume that a diploid parthenogenetic female in exceptional cases may pair with a male of either the species from which it has arisen or some other related species. SZÉKESSY (1937, p. 579—581) has ascertained that the female genital parts of the parthenogenetic curculionids have

remained unchanged. If an egg-cell is fertilised under these circumstances, a triploid parthenogenetic form is of course produced; this naturally presupposes that the fertilising sperm contains an X-chromosome. Such a triploid female might further in the same way give rise to a tetraploid form. It has been pointed out (*e. g.* VANDEL, 1931, p. 316) that polyploid parthenogenetic races do not as a rule cross with diploid bisexual races. SEILER (1927 and 1939), however, has shown that in *Solenobia*, for instance, a cross of this kind may take place, though it cannot produce a permanent triploid race, because of the resultant intersexuality. — As I have found polynucleate spermatocytes in some bisexual species, I would not altogether reject the possibility of diploid gametes contributing to the polyploidy of curculionids. However, as it is not certain whether they develop into functional sperms and even if they do, they contain for the most part sex chromosomes and autosomes in abnormal proportions (cf. DOBZHANSKY, 1937, p. 220), the part they play in this respect seems questionable. I have never found diploid eggs in bisexual curculionids.

Discussing the importance of the crossing of species in the evolutionary process, FEDERLEY (1932, p. 369—382) elucidates the causes that normally prevent the birth of constant species hybrids in animals. He remarks: »Für den Verlauf der Gametogenese eines Bastards sind die Chromosomenverhältnisse von entscheidender Bedeutung». For a regular division of the chromosomes in the meiosis of the hybrids to be possible and for the hybrid to be fertile, it is necessary that the parent species both have the same chromosome number and that, further, there is a complete affinity between their chromosomes. These conditions are seldom fulfilled. Moreover, the gametes of the hybrids must be capable of forming a functional zygote. None of these conditions is, however, necessary in the crosses assumed by me to be the cause of polyploidy in curculionids, since the hybrid is parthenogenetic and its eggs develop without reduction. FEDERLEY (l. c.) also shows that in moth-hybrids the two sexes may occur at quite different times, and thus cannot pair. In the curculionids in question not even this can prevent the survival of the supposed hybrids, since they reproduce parthenogenetically. A change in the time of occurrence might conceivably in these species compel the new form to adapt itself to new ecological conditions, which, for its part, would favour the origin of geographical parthenogenesis. Thus, though in the curculionids studied the parthenogenesis and polyploidy need not have arisen simultaneously,

yet they are not independent of each other. *Parthenogenesis has made polyploidy possible.*

Though it is generally supposed that polyploidy in animals is of no importance for the origin of species, my results seem to show that *in parthenogenetic animals, the egg-cells of which develop without reduction, polyploidy plays a part in the formation of new species* and consequently in the process of evolution itself.

Acknowledgment. — I am greatly indebted to Professor HARRY FEDERLEY, Director of the Institute of Genetics of the University of Helsinki, for the valuable help he has given me in this work.

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ZUR GENETIK VON PHASEOLUS VULGARIS

XV. ÜBER DIE VERERBUNG DER MEHRFARBIG-KEIT DER TESTA

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(With a Summary in English)

EINLEITUNG.

DIE Samen von *Phaseolus vulgaris* zeigen bekanntlich einen ausserordentlichen Reichtum an verschiedenen, erblich bedingten Testafarben. Bisher sind etwa 60 verschiedene Testafarben genetisch analysiert. Zu diesen kommen ferner zahlreiche mehrfarbige und teilfarbige Typen sowie solche mit sog. »Abzeichen».

Die mannigfaltigen Typen von Färbung der Testa können zunächst in drei Hauptgruppen eingeteilt werden, nämlich:

1) Testa farblos,
2) Testa ganzfarbig, und
3) Testa teilfarbig.

Samen mit *farbloser Testa* erscheinen zufolge zwischen dieser und den Keimblättern vorhandener Luft Weiss.

Ganzfarbige Typen haben eine oder mehrere Farben über die ganze Testa verbreitet oder verteilt. Sie lassen sich in zwei scharf gegeneinander abgegrenzte Untergruppen einteilen:

A) Einfarbige,
B) Mehrfarbige.

Bei den *einfarbigen Typen* zeigt die ganze Testa mit Ausnahme des Hilumrandes eine einheitliche Färbung. Die Färbung des Hilumrandes wird (s. LAMPRECHT, 1933, S. 250—252 und 1939 b) durch einen pleiotropen Effekt der die Farbe der Testa im übrigen bedingenden Gene verursacht. Anschliessend an den Hilumrand können begrenzte Zeichnungen, sog. Abzeichen, vorkommen. Zu diesen gehören u. a. Carunculastrich, Corona, Margo und Mikropylenstreifen (s. LAMPRECHT, 1932 b, 1933, 1934 a und 1939 b). Diese Abzeichen werden durch besondere Gene bedingt, sind aber in ihrer Ausbildung überdies vom Vorhandensein gewisser Farbgene in dominanter bzw. rezessiver Form abhängig. Abzeichen können innerhalb aller Gruppen von Fär-

bung der Testa, also auch bei mehrfarbigen und teilfarbigen, auftreten. In vorliegender Arbeit werden sie nicht berücksichtigt.

Die *mehrfarbigen Typen* werden dadurch charakterisiert, dass die Samenschale — abgesehen vom Hilumrand und von ev. Abzeichen — wenigstens zwei Farben aufweist, die mehr oder weniger regelmässig über die ganze Testa verbreitet sind. Bohnensamen, bei denen etwa die Hälfte, ein Drittel usw. eine Farbe, der andere Teil eine andere Farbe aufweist, scheint es nicht zu geben.

Mit Hinblick auf die verschiedene Zeichnung der mehrfarbigen Typen lassen sich diese wie folgt in vier Untergruppen einteilen:

a) *Gestreifte* (gebänderte) Typen;

b) *Homozygot marmorierte* (und überdies stets auch gestreifte) Typen;

c) *Heterozygot marmorierte* Typen;

d) *Bespritzte* Typen.

Die Fig. 2—6 im weiteren Teil der Arbeit zeigen Repräsentanten dieser vier Gruppen.

Eine Unterteilung der mehrfarbigen Typen könnte auch auf Grund der Anzahl verschiedener Farben der Testa versucht werden. Eine solche Einteilung in 2-, 3- usw. farbige Typen wäre jedoch im Vergleich mit der vorstehenden als wenig Aufschluss gebend und unklar zu bezeichnen, da die Zwei-, Drei- usw. Farbigkeit lediglich durch verschiedene Kombination der oben angeführten Gruppen a—d bedingt wird. So sind Samen der Gruppen a) gestreift und c) heterozygot marmoriert stets nur zweifarbig. Homozygot marmorierte Samen, Gruppe b), sind stets dreifarbig (wenn dies auch mitunter bei dunkler gefärbten Samen schwer zu erkennen ist). Die Kombination von b) und c), also gleichzeitig homo- und heterozygot marmoriert, ist vierfarbig, usw. Die vorliegende Arbeit wird sich, wie schon der Titel angibt, ausschliesslich mit der Vererbung der Mehrfarbigkeit befassen.

Die *teilfarbigen Samen* bilden die dritte Hauptgruppe (s. LAMP-RECHT, 1934 b). Bei diesen Typen ist stets ein gewisser Teil der Testa Reinweiss. Der gefärbte Teil der Samenschale hat seinen Ausgangspunkt stets vom Hilumrand. Samen, bei denen die Umgebung des Hilums weiss, andere Teile aber gefärbt sind, scheint es nicht zu geben. Die farbigen Partien auf der Testa dieser Typen zeigen, abgesehen von kleineren Variationen in der Ausbreitung, stets eine symmetrische Anordnung zu beiden Seiten der Längsachse der Samen. Fig. 1 zeigt drei Samen mit verschiedener Teilfarbigkeit. Zu den teilfarbigen Typen

gehören also z. B. nicht Trübrosa auf Weiss gestreifte Samen, denn bei diesen ist die Farbe mehr oder weniger regelmässig über die *ganze* Testa verbreitet. Dieser Typus gehört also zu den ganz- und mehrfarbigen, Gruppe 2 A. Alles, was für die zweite Hauptgruppe, die ganzfarbigen Typen, gesagt worden ist, gilt auch für die teilfarbigen. Es
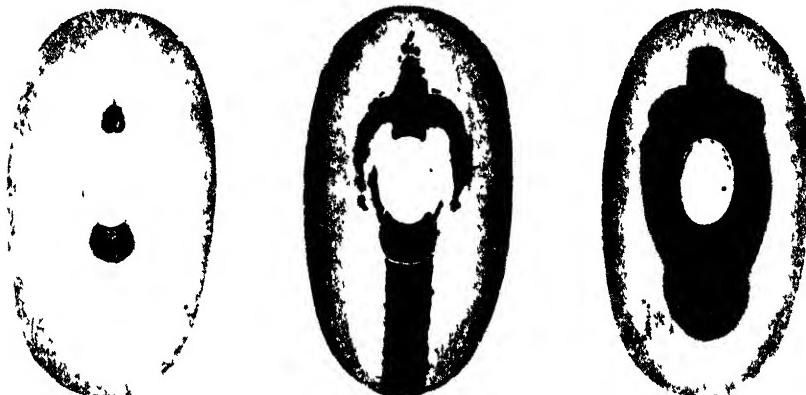

Fig. 1 Drei Samen mit verschiedener Teilfarbigkeit   Links der *bipunctata*-Typus, in der Mitte der *virgarcus*- und rechts der *sellatus*-Typus

können demnach auch diese einfarbig, verschieden mehrfarbig sowie mit verschiedenen Abzeichen auftreten.

DIE VERERBUNG DER STREIFUNG DER TESTA.

Der gestreifte (gebänderte) Samentypus wird dadurch charakterisiert, dass die Testa, seitlich betrachtet, auf hellerem Grunde ein, zwei oder mehrere Streifen (Bänder) in dunklerer Farbe aufweist, die fast stets Unregelmässigkeiten zeigen und bald hier, bald da unterbrochen, zum Teil in kleinere Flecken aufgelöst, bald miteinander mehr oder weniger verflossen sind. Wie Fig. 2 zeigt, verlaufen diese Streifen stets deutlich konzentrisch um das Hilum. Diesen Typus habe ich mehrmals aus Botanischen Gärten unter der Bezeichnung *Dolichos Zebra* bzw. *Phaseolus Zebra* FINGERH. erhalten.˙ Es handelte sich aber stets nur um eine Form von *Phaseolus vulgaris*, denn teils war diese Form ganz mit anderen Rassen von *Ph. vulgaris* übereinstimmend, teils gab sie bei Kreuzung mit solchen ganz fertile Nachkommen.

Die Vererbung der Streifung scheint zuerst von E. v. TSCHERMAK (1912) untersucht worden zu sein. Er kreuzte Flageolet purpurn marmoriert (*MS*) mit Heinrichs Riesen (*mS*), erhielt in F_1 »marmorierte» Samen und in F_2 Spaltung nach 3 Marmoriert : 1 Gestreift. Von

E. v. Tschermak wird also zuerst das Symbol *S* verwendet. Kajanus (1914) findet in einer Kreuzung Dominanz von Marmorierung über Streifung und Dominanz von Streifung über Einfarbigkeit. Tjebbes und Kooiman (1919 und 1921 a, b, c) studierten Kreuzungen zwischen gestreiften (*S*) und einfarbigen (*s*) Samentypen. Sie fanden (1919) in einer Kreuzung zwischen »Haricot de Prague marbré nain» und »Haricot brun clair nain» in F_2 Spaltung nach 1 Schamois Weinrot gestreift : 2 Hellbraun/Schamois marmoriert und Weinrot gestreift : 1 Hellbraun. Je ein Viertel der F_2-Samen entsprach also den Eltern. Mit Hinblick auf die anscheinend monohybride Spaltung nahmen die Verfasser an, dass das Gen *S* die Hellbraune Farbe in Weinrot umwan-
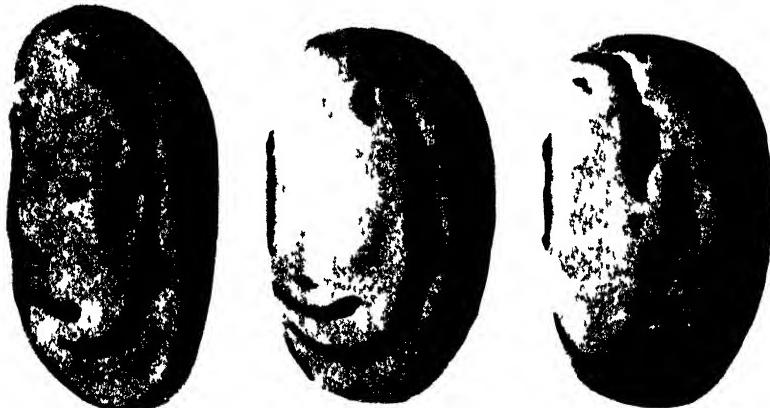
Fig. 2   Drei Samen vom gestreiften Typus (*RS*)   Linker aus L 9, Souvenir de Deuil, die beiden rechten aus »Dolichos Zebra» (= L 145 von *Ph vulgaris*).

delt und diese Farbe überdies nur als Streifen auftreten lässt. — Die Marmorierung war zweifellos auf Heterozygotie in *C* (von Tjebbes und Kooiman mit *B* bezeichnet) zurückzuführen. Diese Verfasser nehmen daher (1921 a) vollkommene Repulsion zwischen *S* und *C* an. Bald darauf (1921 b) studierten sie eine spontane Kreuzung mit Stok-Kievitsboon als Mutter. In dieser trat Streifung auch in anderen Farben, Blauviolett und Braunschwarz auf. Es wurden daher zwei weitere Gene, *Bl* und *Z*, angenommen. *Bl* sollte die roten Streifen in Blauviolette und *Z* in Braunschwarze umwandeln. Das Gen *Bl* ist identisch mit *V*, welche Bezeichnung für dieses Gen schon von Johannsen (1909) verwendet worden ist. *Z* ist kaum sicher zu identifizieren, dürfte aber am ehesten *B* sein. Das Symbol *Z* wurde von E. v. Tschermak (1912) zuerst als Bezeichnung eines Gens für Teilfarbigkeit benutzt.

Nachdem Tjebbes und Kooiman später auch einfarbig »Rote»

Samen (Canadian Wonder) kennen lernten, spricht TJEBBES (1931) von zwei Genen R und S, wobei R die rote Farbe und S die Streifung verursachen soll. Zwischen beiden soll sehr starke Koppelung bestehen und Gleiches soll mit C der Fall sein. TJEBBES berichtet l. c., dass er unter 6500 Pflanzen fünf Umkombinationen zwischen C (= B) und RS und vier zwischen R und S gefunden hat. Die Bezeichnung R für den »Rotfaktor» wurde von TJEBBES gewählt. SHAW and NORTON (1918) sprachen von einer Gelb-Schwarz- und von einer Rot-Serie, die sie durch ein Gen M bzw. M' bedingt erachten. Das Symbol R wurde von mir (LAMPRECHT, 1933) für das Farbgen übernommen, das allein zusammen mit dem Grundgen P die Testafarbe Blass Trübrosa bedingt. Es gibt zusammen mit den verschiedenen anderen Farbgenen von *Phaseolus vulgaris* nicht nur Rote Farbtöne, wie SHAW and NORTON annahmen, sondern Rote, Violette, Bläuliche, Graue Farben und Schwarz.

Für das Verständnis der Wirkung des Gens S ist sein Verhalten zu den verschiedenen Testafarbgenen entscheidend. Und über dieses ist bisher nur sehr wenig bekannt. TJEBBES erwähnt nur das Vorkommen von zwei gestreiften Typen, einen mit Weinroten und einen mit Schwarzblauen Streifen. Im folgenden sollen die Ergebnisse von Kreuzungen mitgeteilt werden, die die Wirkung von S auf die nunmehr gut analysierten Farbgene C, J, G, B, V und R in gewissem Ausmasse klarlegen.

Zur Charakteristik und Bezeichnung der Farben werden benutzt:

RC = Répertoire de Couleurs publié par la Société française des Chrysanthémistes et RENÉ OBERTHUR, Paris 1905.

CS = Color Standards and Nomenclature by ROBERT RIDGWAY, Washington 1912.

FT = Farbentafeln nach OSTWALD, bearbeitet von der Deutschen Werkstelle für Farbkunde.

Kreuzung Nr. 58, Linie 9 × Linie 27. — Linie 27 stammt aus der französischen Wachsbohne De Digoin und hat die Formel *P c J g b v r*; ihre Testafarbe ist in Übereinstimmung hiermit Rohseidengelb (vgl. LAMPRECHT, 1932 a). Linie 9 stammt aus der französischen Brechbohne Souvenir de Deuil; ihre Samen haben die Testafarbe Amethystviolett gestreift auf Rohseidengelbem Grund. Für diese Zeichnung der Testa wird im folgenden stets einfacher geschrieben Amethystviolett S/Rohseidengelb. Fig. 2 zeigt das Aussehen solcher Samen. In typischer Ausbildung entspricht die Farbe der Streifen Améthyste Nr. 197 im RC. Sie zeigt wenig Variation, mitunter ist sie etwas dunkler und

ein wenig bläulicher.   Sie dürfte mit TJEBBES' »striées bleu grisâtre» übereinstimmen.   Die Grundfarbe Rohseidengelb dunkelt schnell nach und wird in wenigen Jahren Abgestorben Laubgelb (RC 321/3).   Damit bekommen auch die Streifen ein anderes Aussehen, ihre Farbe wird mit Bräunlich durchsetzt.

Die auf der ersten Generation dieser Kreuzung erhaltenen Samen waren jenen der einen Elternlinie, Nr. 9, sehr ähnlich.   Sie unterschieden sich nur darin, dass der Rohseidengelbe Grund einen mehr oder weniger deutlichen Anflug von Hell Amethystviolett zeigte.

In der zweiten Generation wurde eine Aufspaltung in fünf verschiedene Testafarben beobachtet.   Tab. 1 zeigt die erhaltenen Individuenzahlen und Farben.   Von diesen sind bisher genetisch analysiert

TABELLE 1.   *Aufspaltung des Bastards PP cc JJ gg bb Vv Rr Ss in F_2 von Kr. 58.*

Testafarbe	Anzahl Pflanzen		D/m für 12:24:12:12:4
	Gefunden	Erwartet	
Amethystviolett *S*/Rohseidengelb......	104	92,44	1,34
Amethystviolett *Ss*/Rohseidengelb mit violettem Anflug	183	184,87	0,17
Eisenhutviolett............................	92	92,44	0,05
Weinrot *S*/Rohseidengelb	87	92,44	0,64
Rohseidengelb	27	30,81	0,71

Rohseidengelb, *P J*, und Eisenhutviolett, *P J V* (s. LAMPRECHT, 1939 a). Da Weinrot *S*/Rohseidengelb die Gene *R* und *S* enthält, haben wir es hier demnach mit einer Spaltung in den drei Genen *V*, *R* und *S* zu tun. Bei einer Dreigenenspaltung sind 8 verschiedene Homozygoten zu erwarten.   Hier wurden nur vier erhalten.   Die fünfte Farbe, Amethystviolett *Ss*/Rohseidengelb mit violettem Anflug, hat sich, wie schon die Schreibweise andeutet, als in *S* heterozygot herausgestellt.   Sie spaltete in F_3 stets in *S*, zum Teil auch in *V*.   Zu den Testafarben mit Streifen (*SS* und *Ss*) ist zu bemerken, dass die Grundfarbe, in vorliegender Kreuzung Rohseidengelb, stets eine schwache Tönung aufweist, die von der Farbe der Streifung herrührt.   So ist der Rohseidengelbe Grund bei Samen mit Weinroten Streifen stets ein wenig Rosa getönt.

Der vollständige Ausfall an vier Genotypen ist hier auf die sehr starke Koppelung zwischen *R* und *S* zurückzuführen.   Mit Hinsicht hierauf ergibt sich für F_2 das folgende Spaltungsschema, das in voller

Übereinstimmung steht mit den in Tab. 1 mitgeteilten Spaltungsergebnissen. Diese zeigen:

1) Starke Koppelung zwischen S und R. Unter den 393 F_2-Indivi-

F_1:
$PP\ cc\ JJ\ gg\ bb\ Vv$
$Rr\ Ss$
Amethystviolett Ss/Rohseidengelb mit amethystviolettem Anflug.

F_2:

$$
\begin{cases}
48\ V \begin{cases}
36\ R \begin{cases} 36\ VRS,\ \text{Amethystviolett } S \text{ und} \\ \qquad\qquad\qquad Ss/\text{Rohseidengelb} \\ 0\ VRs \end{cases} \\
12\ r \begin{cases} 0\ VrS \\ 12\ Vrs\ \text{Eisenhutviolett} \end{cases}
\end{cases} \\[2em]
16\ v \begin{cases}
12\ R \begin{cases} 12\ vRS\ \text{Weinrot } S \text{ und } Ss/\text{Roh-} \\ \qquad\qquad\qquad \text{seidengelb} \\ 0\ vRs \end{cases} \\
4\ r \begin{cases} 0\ vrS \\ 4\ vrs\ \text{Rohseidengelb} \end{cases}
\end{cases}
\end{cases}
$$

Spaltungsschema für F_2 von Kreuzung Nr. 58, L. 9 × L. 27.

duen ist kein einziger Koppelungsbruch zwischen S und R beobachtet worden.

2) Das Gen S begrenzt in homozygoter Form sowohl die Farbenwirkung von R wie von V auf die Streifen. Bei Heterozygotie in S wird die durch V bedingte Farbe nicht vollständig auf die Streifen begrenzt, sie tritt in schwächerer Ausbildung, als deutlich amethystvioletter Anflug auch auf der übrigen Grundfarbe, Rohseidengelb, auf.

3) Auf das Gen J, das die Rohseidengelbe Grundfarbe bedingt, scheint S keinerlei Einfluss zu haben.

Ob das Gen S nur bei Anwesenheit von R in dominanter Form die Streifung bedingt, kann wegen der starken Koppelung mit R einstweilen nicht entschieden werden. Ist dies nicht der Fall, so sollten bei genügend grossem Material auch Individuen mit Samen erhalten werden, die eine Eisenhutviolette Streifung auf Rohseidengelbem Grund zeigen: $J\ V\ S\ r$. Würde S auf Rohseidengelb (J) allein wirken, so sollten Samen mit Rohseidengelber Streifung auf Weiss erhalten werden können, analog wie Trübrosa S/Weiss Samen bekannt sind (z. B. Sorte Heinrichs Riesen).

Kreuzung Nr. 55, Linie 9 × Linie 32. — Linie 9, $P\ J\ V\ R\ S$, wurde in voriger Kreuzung erwähnt, L. 32 stammt aus der französischen Brechbohne Coco marbré und hat die Formel $P\ J\ R\ S$. Ihre Samen sind also demgemäss Weinrot S/Rohseidengelb. F_1 dieser Kreuzung zeigte die Testafarbe Amethystviolett S/Rohseidengelb, aber da S homozygot ist, ohne den Amethystvioletten Anflug auf der Grundfarbe Rohseidengelb, so wie dies in Kr. 58 beobachtet worden ist. In F_2 wurde, wie erwartet, nur monohybride Spaltung im Gen V beobachtet. Es wurden

gefunden 48 Amethystviolett S/Rohseidengelb : 11 Weinrot S/Rohseidengelb. D/m für 3 : 1 = 1,13, also gute Übereinstimmung mit dem theoretisch erwarteten Zahlenverhältnis anzeigend.

Kreuzung Nr. 51, Linie 9 × Linie 29. — L. 9 vgl. Kreuzung Nr. 58.

TABELLE 2. *Die Aufspaltung des Bastards PP Cc Jj gg bb Vv Rr Ss in F₂ von Kreuzung Nr. 51.*

Genenspaltung				Testafarben	Gefunden	Erwartet	D/m
F₂:							
64 CC	48 J	36 V		Veilchenviolett..........	148	135,28	1,18
		12 v		Schamois	50	45,09	0,75
	16 j	12 V		Hell Umbra	31	45,09	2,15
		4 v		Geschwefeltes Weiss...	14	15,03	0,27
128 Cc	96 J	72 V Rr Ss...		Schwarzviolett Ss/Veilchenviolett/Rohseidengelb mit violettem Anflug	255	270,56	1,12
		24 v Rr Ss...		Dunkel Weinrot Ss/-Schamois/Rohseidengelb	101	90,19	1,20
	32 j	24 V Rr Ss...		Dunkel Drab Ss/Hell Umbra/Weiss	88	90,19	0,24
		8 v Rr Ss...		Hell Lila Ss/Geschwefeltes Weiss/Weiss...	32	30,06	0,36
64 cc	48 J	36 V	27 R S	Amethystviolett S/Rohseidengelb	134	101,46	0,12
			9 r s	(Eisenhutviolett)		33,82	
		12 v	9 R S	Weinrot S/Rohseidengelb	49	33,82	0,60
			3 r s	(Rohseidengelb)		11,27	
	16 j	12 V	9 R S	Drab S/Weiss	44	33,82	0,17
			3 r s	Blass Glaucescens	1	11,27	
		4 v	3 R S	Trübrosa S/Weiss	14	11,27	0,27
			1 r s	Reinweiss	1	3,76	

L. 29 stammt aus der französischen Brechbohnensorte de la Chine und hat die Formel *P C j g b v r s.* Der F_1-Generation soll demnach die Formel· *PP Cc Jj gg bb Vv Rr Ss* zukommen. Da Heterozygotie in *C* besteht, sollen die auf F_1 erhaltenen Samen marmoriert und im Zusam-

menhang mit *S* überdies gestreift sein. Dies traf zu. Die Samen zeigten die Farbe Schwarzviolett *S*/Veilchenviolett/Rohseidengelb mit Violettem Anflug. Die in F_2 erhaltenen Resultate sind in Tab. 2 zusammengefasst.

Tab. 2 zeigt, dass die 962 F_2-Individuen sich auf 14 verschiedene Testafarben verteilen liessen. Zwei Farben, Blass Glaucescens und Reinweiss sind allerdings nur mit je einem Individuum vertreten. Unter der Voraussetzung, dass die Koppelung zwischen den drei Genen *C—R—S* so stark wäre, dass es bei vorliegender Individuenanzahl zu keinem Koppelungsbruch kommt, sollten nur 12 verschiedene Testafarben auftreten. Wie aus der Kolonne Genenspaltung in der Tabelle hervorgeht, repräsentieren gerade die beiden vorhin erwähnten Testafarben mit je nur einem Individuum einen Koppelungsbruch zwischen einerseits *C* und andererseits *R—S*. Der Crossingoverprozent beträgt hier noch nicht 0,5 %. Erwähnt sei, dass bei der Berechnung der Werte für D/m auf die genannten beiden abweichenden Individuen keine Rücksicht genommen worden ist; die hierdurch bedingte zahlenmässige Änderung ist völlig belanglos für die Beurteilung der Ergebnisse.

In den anderen Testafarben, die durch einen solchen Koppelungsbruch entstehen könnten, nämlich Eisenhutviolett und Rohseidengelb sowie Gestreift auf den Farben Veilchenviolett, Schamois, Hell Umbra und Geschwefeltes Weiss, wurde kein Individuum erhalten. Die Spaltungsergebnisse zeigen ferner, dass zwischen *R* und *S* kein einziger Koppelungsbruch hat festgestellt werden können. Ganz- und einfarbige Samen mit *R*, wie z. B. Pflaumenviolett usw., fehlen demnach vollständig. Streifung konnte daher nur beobachtet werden bei *Cc*- und *cc*-Individuen. Die in *C* heterozygoten Pflanzen sind daher auch gleichzeitig stets in *R* und *S* heterozygot. Entsprechend dieser Konstitution tritt die Streifung auf *Cc*-marmoriert auf. Bei dieser sog. Heterozygotmarmorierung entsprechen die dunkleren Flecken dem Genotypus mit *CC*, der hellere Grund demjenigen mit *cc*, aber mit im übrigen gleicher Konstitution für die Testafarbe. Die Farbe der Streifen ist bei diesen Typen, wie die Namen angeben, stets eine etwas dunklere. Von der durch Heterozygotie in *R* bei ganzfarbigen *R*-Samen bedingten schwachen, hauptsächlich um das Hilum erkennbaren Marmorierung (vgl. LAMPRECHT, 1934 c) ist bei diesen gestreiften Typen nichts wahrzunehmen.

Kreuzung Nr. 51 hat die genetische Analyse von zwei weiteren gestreiften Testafarben erbracht, nämlich: Drab *S*/Weiss, *P V R S*, und Trübrosa *S*/Weiss, *P R S*. Da die Drab-Streifung auf Reinweissem

Grunde auftritt, besagt dies, dass das Gen S auch die durch V bedingte Farbe Blass Glaucescens auf die Streifen vereinigt. Gleiches ist schon in Kr. 58 bei der Farbe Amethystviolett S/Rohseidengelb festgestellt worden.

Die Farbe der Drab Streifen entspricht — wenn typisch — im CS Drab bis Hair-Brown, XLVI, 17‴, 0—1, im RC am nächsten Fischotterfarbig, 354/3—4. Die Farbe zeigt, wie fast alle mit V in ihrer Konstitution, eine beträchtliche Variation; Aschfarbig, RC 358/2—4 über Fischotterfarbig bis zu Neutraltinte, RC 361/2—4. Bei ersterer tritt die Wirkung von V stärker zutage, bei letzterer die von R. Die Farbe der Trübrosa Streifen entspricht im CS etwa Pinkish Vinaceous, XXVII, 5″, d, im RC Rötlich Lila, 179/1. Nicht selten ist diese Farbe etwas blasser und weniger rein Rötlich Lila, mehr Schmutzig Rosa; doch ist die Variation nicht bedeutend.

TABELLE 3. *Die Aufspaltung des Bastards PP cc jj gg bb Vv Rr Ss in F_2 von Kreuzung Nr. 142.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
F_2:	12 V	9 RS ...	Drab S/Weiss	540	565,31	1,61
		3 rs ...	Blass Glaucescens	208	188,44	1,58
	4 v	3 RS ...	Trübrosa S/Weiss	195	188,44	0,53
		1 rs ...	Reinweiss	72	62,81	1,20

Kreuzung Nr. 142, Linie 107 × Linie 118. — Linie 107 stammt aus einer spontanen Kreuzung in Flageolet Wachs. Ihre Konstitution hinsichtlich Testafarbe ist $P c j g b V r$. In Übereinstimmung hiermit zeigten die Samen die Farbe Blass Glaucescens. Linie 118 stammt aus der Brechbohnensorte Heinrichs Riesen. Sie hat die Formel $P c j g b v R S$, also der Farbe Trübrosa S/Weiss entsprechend. Die auf der F_1-Generation erhaltenen Samen zeigten die Farbe Drab S/Weiss, also eine schon aus voriger Kreuzung bekannte Testafarbe.

Die in F_2 erhaltenen Spaltungsergebnisse sind in Tab. 3 wiedergegeben. Sie zeigen das erwartete Resultat. Infolge der starken Koppelung zwischen R und S wurden nur vier Testafarben gefunden, die in dem bifaktoriellen Verhältnis 9 : 3 : 3 : 1 auftraten. Die zahlenmässige Übereinstimmung ist durchweg gut. Neue Testafarben sind hier nicht aufgetreten. Die Ergebnisse bestätigen nur die bereits früher gefundenen. Unter den 1015 F_2-Individuen konnte nicht ein einziger

Koppelungsbruch zwischen den beiden Genen R und S festgestellt werden.

Kreuzung Nr 367, Linie 190 $\times$ *Linie 201.* — Linie 190 stammt von einer durch spontane Kreuzung in der schwedischen Braunen Bohne Apollo aufgetretenen Pflanze. Die Samen dieser Linie zeigen die Testafarbe Mahagonibraun S/Rohseidengelb. Linie 201 stammt aus meiner Kreuzung Nr. 12 (s. LAMPRECHT, 1932 a) und hat Reinweisse Samen, sowie die Konstitution P mit allen Farbgenen in rezessiver Form. Die Formel von Linie 190 hat sich erst aus vorliegender Kreuzung ergeben, sie ist $P\,c\,J\,G\,b\,v\,R\,S$.

TABELLE 4. *Die Aufspaltung des Bastards PP cc Jj Gg bb vv Rr Ss in F_2 von Kreuzung Nr. 367.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
F_2:	48 J	36 G { 27 RS	Mahagonibraun SS/- Rohseidengelb.........	148	143,86	0,38
		36 G { 9 rs 	Mahagonibraun Ss/- Hell Maisgelb	287	287,72	0,05
		9 rs	Maisgelb	152	143,86	0,73
		12 g { 9 RS	Weinrot S/Rohseiden- gelb....................	136	143,86	0,71
		12 g { 3 rs	Rohseidengelb	48	47,95	0,01
	16 j	12 G { 9 RS	Hell Braunrot S/Speck- weiss	135	143,86	0,80
		12 G { 3 rs	Speckweiss	61	47,95	1,95
		4 g { 3 RS	Trübrosa S/Weiss......	43	47,95	0,73
		4 g { 1 rs	Reinweiss................	13	15,98	0,76

Da die eine Elternlinie, Nr. 201, sämtliche Farbgene rezessiv hat, und beide in c rezessiv sind, wäre zu erwarten gewesen, dass die auf F_1 erhaltenen Samen in der Farbe ganz mit dem des einen Elters, L. 190, übereinstimmen sollten. Die durch die Gene J und G bedingten Farben zeigen ja sonst vollkommene Dominanz. Dies war indessen nicht der Fall. Die Farbe dieser Samen war nicht Mahagonibraun S/Rohseidengelb sondern Mahagonibraun Ss/Hell Maisgelb. Die Grundfarbe war Maisgelb, der Formel $P\,J\,G$ entsprechend, aber etwas heller. Die Erklärung dieser Erscheinung gibt die F_2-Generation. Die Spaltung in dieser ist in Tab. 4 wiedergegeben.

Tab. 4 nimmt 9 verschiedene Testafarben auf. Wie ersichtlich

fand eine Spaltung in 4 Genen, *J*, *G*, *R* und *S*, statt. Zwischen *R* und *S* konnte auch in dieser Kreuzung mit insgesamt 1023 Individuen kein einziger Koppelungsbruch beobachtet werden. Es verbleibt also nur Spaltung in drei Genen, da *R S* als ein einziges Gen zu wirken scheint. In diesen drei Genen wird normale Spaltung, dem Verhältnis 27 : 9 : 9 : 9 : 3 : 3 : 3 : 1 entsprechend, gefunden. Die gefundenen Individuenzahlen stimmen gut mit den theoretisch erwarteten überein; die Werte für D/m variieren zwischen 0,01 und 1,95.

Drei neue, bisher nicht analysierte Testafarben wurden konstatiert: Mahagonibraun *S*/Rohseidengelb, Mahagonibraun *Ss*/Hell Maisgelb und Hell Braunrot *S*/Speckweiss. Die Mahagonibraunen Streifen entsprechen im CS Mahogany Red, II, 7, k, im RC Mahagonibraun, 335/1—2. Die Variation dieser Farbe ist ziemlich gering, mitunter ist sie heller. Die Farbe Mahagonibraun *Ss*/Hell Maisgelb beruht, wie schon die Schreibweise *Ss* andeutet, auf Heterozygotie im Gen *S*. Damit folgt natürlich auch stets Heterozygotie im Gen *R*. Die Resultate in F_3 bestätigten dies. Solche Samen spalteten nämlich stets wenigstens in *S* und *R*. Hieraus kann der Schluss gezogen werden, dass *S* auch das Gen *G* beeinflusst. *P J G*-Samen sind Maisgelb, *P J G R S*-Samen sind nicht Rot gestreift auf Maisgelb, sondern Mahagonibraun gestreift auf Rohseidengelb. Die Grundfarbe entspricht also der Formel *P J* und nicht *P J G*. Der durch *G* bedingte Farbeneffekt wird demnach durch *S* auf die gleichen Stellen der Samen, d. h. auf die Streifen, begrenzt wie dies hinsichtlich *R* der Fall ist. Bei Heterozygotie in *S* ist diese Wirkung indessen nur schwach zutagetretend, der Grund der Samenschale ist dann Hell Maisgelb, sowie dies schon in F_1 beobachtet worden ist. Diese Erscheinung bildet ein Gegenstück zu den Samen der Konstitution *P c J V R S*. Diese zeigten (s. Kreuzung Nr. 58) die Farbe Amethystviolett *S*/Rohseidengelb, aber bei Heterozygotie in *S* war die Grundfarbe Rohseidengelb mit violettem Anflug. Durch *SS* wird also sowohl die Farbwirkung von *G* (bei Anwesenheit von *J*) wie von *V* auf die Streifen begrenzt, aber bei Heterozygotie ist diese Wirkung nur eine teilweise, denn zum Teil kommt der Effekt dieser beiden Gene dann auch auf der Grundfarbe zur Geltung. Die zweite in vorliegender Kreuzung neu erwähnte Farbe ist Hell Braunrot *S*/Speckweiss. Die Hell Braunrote Farbe der Streifen entspricht im CS Terra Cotta bis Ocker Red, XXVII, 5″ i—XXVIII, 7″; bei besonders guter Ausbildung kann die Farbe Deep Corinthian Red, XXVII, 3″ i, erreichen. Im RC stimmt sie am besten mit Blutrötlich Braun, 337/1, überein; jedoch ist die Farbe gewöhnlich etwas rötlicher. Die Grundfarbe Speckweiss hat hier ebenso

wie bei Weinrot S/Rohseidengelb gewöhnlich einen schwach rötlichen Anflug. Ein weiterer Einfluss von S ist bei der hier sehr hellen Grundfarbe Speckweiss (G) nicht wahrzunehmen.

Ein Rückblick auf die Resultate der vorhin besprochenen fünf Kreuzungen ergibt folgendes. Es wurde die genetische Konstitution folgender gestreifter Testafarben festgestellt:

Trübrosa S/Weiss, $P\ c\ j\ g\ b\ v\ R\ S$,

Weinrot S/Rohseidengelb, $P\ c\ J\ g\ b\ v\ R\ S$,

Drab S/Weiss, $P\ c\ j\ g\ b\ V\ R\ S$,

Hell braunrot S/Speckweiss, $P\ c\ j\ G\ b\ v\ R\ S$,

Amethystviolett S/Rohseidengelb, $P\ c\ J\ g\ b\ V\ R\ S$,

Amethystviolett Ss/Rohseidengelb mit violettem Anflug, $P\ c\ J\ g\ b$ $V\ R\ Ss$,

Mahagonibraun S/Rohseidengelb, $P\ c\ J\ G\ b\ v\ R\ S$,

Mahagonibraun Ss/Hell Maisgelb, $P\ c\ J\ G\ b\ v\ R\ Ss$.

In bezug auf die von TJEBBES (1931) aufgestellte Koppelungsgruppe C—R—S wurde folgendes festgestellt. C—R zeigten sehr starke Koppelung. Unter 962 Individuen (F_2) wurden 2 Koppelungsbrüche gefunden. TJEBBES hat hierfür unter 6500 Individuen 5 Koppelungsbrüche beobachtet. Zwischen R und S konnte von mir unter den hier mitgeteilten F_2-Generationen mit insgesamt 3552 Individuen kein einziger Koppelungsbruch festgestellt werden. Hier ist noch zu erwähnen, dass die genannten Kreuzungen auch in F_3 und F_4 sowie weitere 12 Kreuzungen studiert worden sind, in denen R und S spalteten. Dieses Material umfasst im ganzen etwa 16.000 Individuen. Aber im ganzen Material ist kein einziger sicherer Fall von Koppelungsbruch beobachtet worden. Es führt dies auf den Gedanken, dass es sich nur um ein einziges Gen handeln könnte, dass die rote Streifung der Testa bedingt, nämlich das Gen S, sowie dies von TJEBBES und KOOIMAN (1921 a) zuerst angenommen worden ist. Und dass das Gen R, das u. a. mit C die Farbe Hell Lila bedingt, nichts mit S zu tun hat. Ist dem so, dann wären die von TJEBBES (1931) mitgeteilten vier Koppelungsbrüche zwischen R und S unter 6500 Individuen anzuzweifeln und vielleicht auf spontane Kreuzung oder andere jetzt nicht mehr kontrollierbare Zufälligkeiten zurückzuführen.

Klarlegend werden hier Kreuzungen sein, in denen Linien mit einfarbig rötlichen Samen mit solchen mit rot gestreiften gekreuzt werden. Ist S ein Gen, das nur Streifung der Testa bedingt, und die Wirkung

des Gens R also auf die Streifen begrenzt, dann müssen die auf F_1 solcher Kreuzungen erhaltenen Samen irgendwelche Rötliche Streifung, aber gleichzeitig keinen Roten Grund aufweisen. Eine solche Kreuzung, Nr. 317, wurde ausgeführt und bisher in F_1 untersucht. Die Eltern waren: Linie 118 aus Heinrichs Riesen, $P R S$, und Linie 165 aus meiner Kreuzung Nr. 49 (s. LAMPRECHT, 1935), $P C R s$. Die Testafarbe der ersteren war also Trübrosa S/Weiss, die der letzteren Hell Lila. Die auf der F_1-Generation erhaltenen 1310 Samen zeigten nun alle die Farbe Dunkel Trübrosa gestreift (S) auf Hell Lila marmoriert (Rr) auf Weiss. Die Anwesenheit von S hatte hier demnach keinen Einfluss auf R, den R gibt ohne S in heterozygoter Form zusammen mit C Hell Lila marmoriert auf Geschwefeltes Weiss. Und S hat ja laut oben mitgeteilter Kreuzung Nr. 51 (Tab. 2) gerade zusammen mit R und C ($Cc Rr Ss$) die Testafarbe Hell Lila Ss/Geschwefeltes Weiss/Weiss verursacht. Von R-Marmorierung war hier also nichts wahrzunehmen, sondern die rote Farbe war durch S nur auf die Streifen lokalisiert. Dies Ergebnis dürfte mit grosser Wahrscheinlichkeit zu dem Schluss berechtigen, dass das Gen R zusammen mit dem Grundgen P die Farbe einfarbig Trübrosa, S dagegen zusammen mit P Trübrosa gestreift auf Weiss bedingt. Mit dieser Annahme sind jedenfalls die gefundenen Resultate zu erklären. Der letztere Typus Trübrosa S/Weiss wurde u. a. in den Kreuzungen Nr. 51, 142 und 367 erhalten. Dieser Testafarbe entspräche dann die Formel $P c r S$. Gleiches gilt für die Linien Nr. 118 und 130 aus Heinrichs Riesen. Linie 165 mit der Testafarbe Hell Lila hätte dann die Formel $P C R s$. Und bei Kreuzung von L. 118 mit L. 165 ergibt sich dann: $P Cc Rr Ss$, also der Testafarbe Dunkel Trübrosa S/Hell Lila Rr/Weiss entsprechend. Die Marmorierungen von Cc und Rr fallen nämlich in ihrer Zeichnung zusammen. Mitunter kann jedoch, bei schwächerer Ausbildung der Rr-Marmorierung, was nicht selten vorkommt, auch noch etwas von der durch Cc bedingten Marmorierung Geschwefeltes Weiss/Weiss wahrgenommen werden. Klaren Bescheid über das Verhältnis von R zu S werden die weiteren Generationen geben.

DIE VERERBUNG DER HOMO- UND HETEROZYGOTEN MARMORIERUNG.

Schon E. v. TSCHERMAK (1901, 1902, 1904), der als erster die Vererbung der Marmorierung studierte, fand sowohl homozygote wie heterozygote Marmorierung. Im ersten Fall wurde also monohybride

Spaltung nach 3 Marmoriert : 1 Einfarbig, im letzteren 2 Einfarbig : 2 Marmoriert festgestellt. Diese beiden Typen von Vererbung der Marmorierung· (der eine oder beide) wurden seither von einer Reihe von Forschern beobachtet (EMERSON, 1904; SHULL, 1907, 1908; EMERSON, 1909 a, 1909 b; E. v. TSCHERMAK, 1912; KAJANUS, 1914; SHAW and NORTON, 1918; TJEBBES und KOOIMAN, 1919 a, 1919 b; KOOIMAN, 1920; SIRKS, 1920, 1922 a, 1922 b; KRISTOFFERSON, 1924; MIYAKE, 1930, LAMPRECHT, 1932 a, 1933, 1935, 1936, 1939 a und PRAKKEN, 1934). Im folgenden werden nur solche Arbeiten berücksichtigt, die für das Verständnis der bisher aufgestellten Hypothesen von Belang sind.

SHULL (l. c.) nahm ein Marmorierungsgen M an, das im heterozygoten Zustand Marmorierung bedingen soll. Dieses Gen sollte natürlich auch in weissen Samen (p) latent vorhanden sein können, wodurch dann bei Kreuzung mit einfarbigen Linien die F_2-Spaltung 6 Marmoriert : 6 Einfarbig : 4 Weiss resultierte.

EMERSON (1909 a) nahm zuerst M als Gen für die konstante Marmorierung an und X als Gen, das in heterozygoter Form Marmorierung bedingte. Kurz darauf machte sich bei EMERSON (1909 b) der Wunsch geltend die Vererbung beider Arten von Marmorierung durch eine gemeinsame Hypothese genetisch unter einen Hut zu bringen. Beeinflusst durch SPILLMAN wurde (l. c.) folgende EMERSON—SPILLMANsche Hypothese aufgestellt. Es bestehen zwei Gene, Y und Z, die absolut gekoppelt sind und Marmorierung tritt nur auf, wenn beide dominant anwesend sind. Konstant marmorierte Samen wären danach $YY\,ZZ$, einfarbige $YY\,zz$, $yy\,ZZ$ oder $yy\,zz$. Diese Hypothese steht anscheinend sowohl mit der Spaltung von konstant marmorierten wie von heterozygot marmorierten im Einklang. Nach Kreuzung der beiden einfarbigen Typen $YY\,zz \times yy\,ZZ$ erhält man $Yy\,Zz$, also — da beide Gene in dominanter Form vorhanden sind — marmorierte F_1. Die Nachkommen dieser spalten zufolge der absoluten Koppelung nach 1 $YY\,zz$: 2 $Yy\,Zz$: 1 $yy\,ZZ$. Wird $YY\,zz$ mit $YY\,zz$ oder mit $yy\,zz$, bzw. $yy\,ZZ$ mit $yy\,ZZ$ oder mit $yy\,zz$ gekreuzt, werden stets nur Kombinationen mit entweder allein YY oder ZZ, demnach einfarbige Typen erhalten, was gleichfalls oft zu konstatieren ist. Schliesslich ist laut dieser Hypothese nach Kreuzung von $YY\,ZZ$ (konstant marmoriert) mit jedem der drei Genotypen $YY\,zz$, $yy\,ZZ$ und $yy\,zz$ Spaltung im Verhältnis 3 Marmoriert : 1 Einfarbig zu erwarten, was tatsächlich zutrifft. Rassen der Formel $yy\,zz$ haben bisher indessen nicht angetroffen werden können. Die Konstitution $yy\,zz$ wäre ja leicht festzustellen, denn eine solche Rasse müsste sowohl mit $YY\,zz$ wie mit $yy\,ZZ$ (die bei

Kreuzung miteinander beständig spaltende marmorierte geben) einfar-
bige Nachkommen liefern.

E. v. TSCHERMAK (1912) stellte eine neue Hypothese auf, nach der
zwischen dem Marmorierungsgen M und dem Grundgen für Testafär-
bung A entweder »Assoziation» oder »Dissoziation» bestehen können
soll. Wenn M mit A assoziiert ist, soll hierdurch die Marmorierung
zustande kommen. Dieser Wirkung von M auf A soll eine lokale Hem-
mung der Pigmentbildung (= Marmorierung) entsprechen. Bei Disso-
ziation soll es keine Marmorierung geben, jedoch soll diese dann durch
die Heterozygotie ausgelöst werden (l. c. S. 186).

SHAW and NORTON (1918) sowie MIYAKE (1930) haben sich der
EMERSON—SPILLMANschen Hypothese angeschlossen. SHAW and NOR-
TON scheinen als erste das Vorkommen von doppelter Marmorierung
festgestellt zu haben. Sie teilen die Testafarben in zwei Serien ein, eine
Rotserie, bei der diese rötliche Farben aufweisen und eine Gelb-Schwarz-
Serie mit gelben-braunen-schwarzen Farben. Die doppelt marmorier-
ten Samen wurden erhalten, wenn eine konstant marmorierte Rasse
der Rotserie mit gewissen einfarbigen Rassen der Gelb-Schwarz-Serie
gekreuzt wurden.

KOOIMAN (1920) findet, dass beständig spaltende Marmorierung
auf Heterozygotie im Farbgen C (von ihm als B bezeichnet, welches
Symbol bereits früher vergeben war; s. LAMPRECHT, 1932 a) zurück-
zuführen ist. In seiner später erschienenen Monographie (1931, S. 371)
fasst er seine Ansicht über die Vererbung verschiedener Marmorierung
mit Hinblick auf bis dahin Bekanntes folgendermassen zusammen:

1. True breeding mottling is the effect of a mottling factor M.
This mottling factor acts with most pigments.

2. The mottling factor may be latent in white-seeded races, and
besides in buff (= chamois)-seeded ones.

3. Inconstant mottling is caused by the heterogenous condition of
one of the brown chromogenous factors, B.

KRISTOFFERSON (1924) nimmt zu diesen Hypothesen keine be-
stimmte Stellung ein, hält sie aber für keineswegs voll klarlegend. In
seinen Formeln verwendet er die Gene Y und Z von EMERSON. Gleich-
wie SHAW and NORTON stellt er das Vorkommen von doppelt marmo-
rierten Samen fast, auf denen also zweierlei verschiedene Marmorierung
vereint ist. F_2 wurde irrtümlicherweise nicht pflanzenweise beurteilt.
Aber in F_3 findet er Spaltung nach 9 doppelt marmoriert : 3 einfach
marmoriert : 3 einfarbig : 1 einfarbig.

LAMPRECHT (1932 a, 1933, 1935, 1936, 1939 a) veröffentlichte ein

umfangreiches Material, das die Bedingtheit der beständig spaltenden Marmorierung durch Heterozygotie in den Genen *C* und *R* (je für sich wie auch zusammen) klarlegt. Nunmehr sind sämtliche Kombinationen der Farbgene *J, G, B, V* sowie der Modifikationsgene *Vir, Och* und *Flav* in ihrem Verhalten zu *CC, cc* und *Cc* untersucht (s. LAMPRECHT, 1939 und 1940). Für die *Cc*-heterozygoten, marmorierten Farben gilt durchweg die Regel, dass die dunkleren Flecken der Samen die dem Genotypus mit *CC*, der hellere Grund die dem im übrigen gleichen Genotypus aber mit *cc* entsprechende Farbe aufweisen. Ganz Analoges gilt für die *Rr*-heterozygoten Samen (soweit diese bisher studiert worden sind).

PRAKKEN (1934) beschäftigt sich recht eingehend mit der Vererbung der Marmorierung und erörtert die bis dahin vorliegenden Hypothesen. Er stellt dann eine eigene Hypothese auf (l. c. S. 226—228), laut der beide Arten von Marmorierung durch zwei absolut gekoppelte Gene *C* und *M* bedingt werden. Diese »neue Hypothese» ist aber im Prinzip ganz gleich der EMERSON—SPILLMANschen; der einzige Unterschied besteht darin, dass PRAKKEN die Symbole *Y* und *Z* von EMERSON durch *C* und *M* ersetzt hat. (Man vergleiche hierzu PRAKKENs Spaltungsfiguren l. c. S. 223 oben und S. 227, die den Ersatz von *Y Z* durch *C M* klar dartun.) PRAKKEN bezieht also ganz einfach das eine Gen EMERSONs (z. B. *Y*) auf das nunmehr sehr gute analysierte Farbgen *C* und *Z* auf *M*. Im Zeitpunkte der Veröffentlichung EMERSONs (1909 b) war die Wirkung von *C* noch unbekannt sowie eine klare Farbgenanalyse von *Phaseolus vulgaris* noch nicht vorliegend.

Ein Rückblick auf bisher über Vererbung von Marmorierung bekannte Tatsachen und Hypothesen ergibt folgendes. Einwandfrei klargelegt erscheint, dass — bei Dominanz in den Grundgenen für Testafarbe *P* und *Gri* — durch Heterozygotie im Farbgen *C* bzw. *R* (oder auch beiden zusammen) Marmorierung bedingt wird. Diese Marmorierung entspricht der in Fig. 3 wiedergegebenen Zeichnung. Sie besteht stets aus *zwei* Farben, einer helleren Grundfarbe, die dem Genotypus mit *cc* bzw. *rr* entspricht, und den dunkleren Flecken, dem gleichen Genotypus aber mit *CC* bzw. *RR* entsprechend. (Wenn die Samen überdies das dominante Farbgen *V* enthalten, so ist die Grundfarbe heller, d. h. die Wirkung von *V* ist hier schwächer zutage tretend, als bei entsprechenden einfarbigen Typen.) Eine zweite Feststellung ist die, dass die konstante (homozygote)

Marmorierung über einfarbig dominiert, also in Kreuzungen mit einfarbigen Spaltung nach 3 marmoriert : 1 einfarbig zeigt.

Eine dritte Feststellung schliesslich ist, dass bei Kreuzung zwischen konstant marmorierten mit gewissen einfarbigen Samentypen doppelte Marmorierung resultiert (SHAW and NORTON, 1918; KRISTOFFERSON, 1924). Die Doppelmarmorierten zeigen dann u. a. eine Spaltung nach 9 doppelt marmoriert : 3 einfach marmoriert : 3 einfarbig : 1 einfarbig.
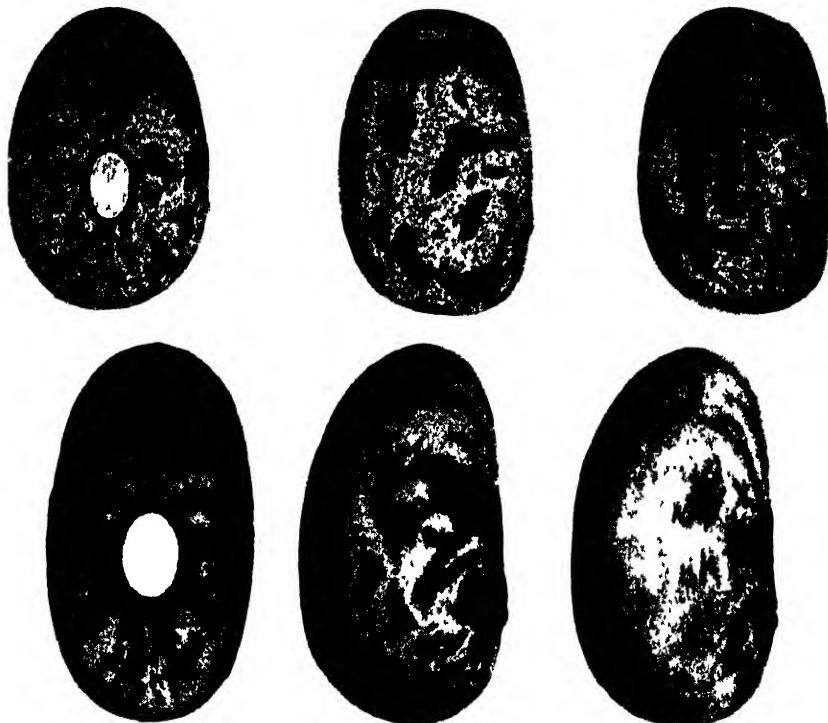
Fig. 3. Sechs Samen mit Heterozygotmarmorierung, die oberen drei Cc, die unteren Rr.

Weitere experimentelle Tatsachen scheinen nicht bekannt zu sein. Da es sich hier in erster Linie um die Beziehungen zwischen konstanter und heterozygoter Marmorierung handelt, erscheinen zwei Fragen von grösstem Interesse: 1) Zeigen diese beiden Typen von Marmorierung dieselbe Zeichnung, dasselbe Muster auf der Samenschale? und 2) Gibt es konstante und heterozygote Marmorierung in gleichen Testafarben? — Und auf diese beiden fundamentalen Fragen für die Beurteilung eines genetischen Zusammenhanges zwischen den beiden Arten

von Marmorierung geben die bisher vorliegenden Untersuchungen entweder gar keinen oder höchstens unklaren Bescheid.

Was die Hypothesen betrifft um beide Arten von Marmorierung genetisch unter einen Hut zu bringen (EMERSON—SPILLMAN, E. v. TSCHERMAK, PRAKKEN), so kann E. v. TSCHERMAKs Hypothese über »Assoziation» und »Dissoziation» zwischen zwei Genen ohne weiters abgelehnt werden; sie ist mit unserer jetzigen Auffassung der Wirkung und des genetischen Verhaltens von Genen und ihren Allelen unvereinbar. Die Hypothesen von EMERSON—SPILLMAN und von PRAKKEN sind, wie bereits erwähnt, im Prinzip gleich. Beide nehmen zwei absolut gekoppelte Gene an und nur bei Dominanz in beiden soll Marmorierung auftreten. Erstere benutzen hierfür die Symbole Y und Z, letzterer C und M; in beiden Fällen werden diese als Marmorierungsgene aufgefasst. Der einzige, aber für die Frage nach der Vererbung der Marmorierungen belanglose Unterschied ist der, dass EMERSON keinem seiner Gene Y und Z eine bestimmte Farbwirkung zuschreibt, während dies bei PRAKKEN der Fall ist (er benutzt mein Gen C für Geschwefeltes Weiss). Laut diesen beiden Hypothesen ergeben sich dann die beiden, bereits vorstehend mit Formeln erläuterten, möglichen Spaltungsverhältnisse 3 marmoriert : 1 einfarbig sowie 1 einfarbig : 2 heterozygot marmoriert : 1 einfarbig.

Gegen diese beiden Hypothesen können nun schon auf Grund der bisher bekannten experimentellen Feststellungen folgende Einwände gemacht werden:

1) Das Auftreten von doppelt marmorierten Typen, wie es von SHAW and NORTON (1918) und KRISTOFFERSON (1924) festgestellt worden ist, kann auf Grund dieser Hypothesen weder erwartet noch mit denselben erklärt werden. Diese gestatten nur das Auftreten von konstanter Marmorierung, $YY\,ZZ$, von 3 : 1 spaltender solcher, $YY\,Zz$ und $Yy\,ZZ$ und von heterozygoter, beständig 1 : 2 : 1 spaltender, $Yy\,Zz$. Und diese sind nicht doppelt sondern einfach marmoriert. Da es also sowohl einfache wie doppelte Marmorierung gibt, kann für beide nicht dieselbe genetische Grundlage angenommen werden. Zumindestens müssten diese Hypothesen hierfür erweitert werden.

2) Laut der EMERSON—SPILLMANschen Hypothese sollen einfarbigen Samen die drei Formeln $YY\,zz$, $yy\,ZZ$ und $yy\,zz$ zukommen können. Es ist nun auffallend, dass bisher niemals Samen der Konstitution $yy\,zz$ angetroffen worden sind. Die Spaltungsresultate aller Forscher sind nur durch $YY\,zz$ und $yy\,ZZ$ erklärbar. In analoger

Weise hebt PRAKKEN für seine Hypothese hervor (1934, S. 227), dass
Samen der Konstitution *cc mm* bisher noch nicht angetroffen sind.

3) Sehr auffallend ist schliesslich, dass in den zahlreichen seit 1900
auf diesem Gebiete erschienenen Arbeiten auch nicht ein einziger Fall
bekannt geworden ist, wo konstante und beständig spaltende Marmo-
rierung die gleichen Testafarben gezeigt hätten. Zur Erklärung dieser
Erscheinung müsste angenommen werden, dass *YY ZZ* andere Farben
bedingt als *Yy Zz* bzw. *CC MM* als *Cc Mm*. Diese Genkonstitutionen
sollten also auch auf die anderen Farbgene einen verschiedenen Ein-
fluss ausüben. Oder schliesslich: Es sind im übrigen gleiche Genotypen
mit diesen beiden alternativen Konstitutionen bisher niemals erhalten
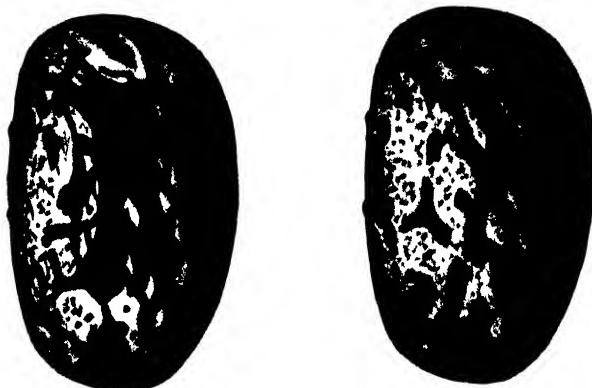
Fig. 4. Zwei konstant (= homozygot) marmorierte Samen. Linker aus L. 47, Huish
Beauty, rechter aus L. 72, Early Prolific.

worden. Eine bei der jetzt sehr weit vorgeschrittenen Genanalyse der
Testafarben allerdings mehr als höchst unwahrscheinliche Annahme.

4) Eine Untersuchung der konstant marmorierten Typen zeigt, dass
die Testa dieser stets sowohl Marmorierung wie Streifung aufweist
(vgl. Fig. 4). Diesbezüglich wurde von mir ein sehr grosses Material
studiert. Alle erreichbaren marmorierten Handelssorten und viele
Tausende aus Kreuzungen erhaltene Individuen. Bei stärkerer, dunkler
Färbung ist es mitunter nicht möglich, diesbezüglich auf Grund okulä-
rer Besichtigung allein Gewissheit zu erlangen. Die Samen können
dann nur marmoriert erscheinen. Aber bei Kreuzung mit heller ge-
färbten Linien hat sich stets herausgestellt, dass auch Streifung vor-
handen war. Diese Erscheinung sollte auch früheren Forschern auf
diesem Gebiete nicht ganz unbekannt geblieben sein. Sie spricht be-
stimmt dafür, dass die Ausbildung von konstanter Marmorierung von

der Anwesenheit des Gens *S* in dominanter Form (Streifung) abhängig ist. Die weiter unten mitgeteilten Kreuzungen werden dies bestätigen.

Ausgehend von der Annahme, dass die genetische Grundlage der konstanten Marmorierung nichts mit der durch *Cc* (bzw. *Rr*) bedingten heterozygoten Marmorierung zu tun hat, habe ich in einer früheren Arbeit (LAMPRECHT, 1933, S. 313) die Möglichkeit eines bindenden Beweises hierfür in der Spaltung nach Kreuzung von *MM CC*-Linien mit *mm cc*-Linien zu finden gesucht. Die Aufspaltung des in Frage stehenden Bastards *Mm Cc* sollte dann in F_2 folgendes Verhältnis geben: 1 *MM CC* : 2 *MM Cc* : 1 *MM cc* : 2 *Mm CC* : 4 *MM Cc* : 2 *Mm cc* : 1 *mm CC* : 2 *mm Cc* : 1 *mm cc*. Da alle Genotypen mit *M* und *Cc* marmoriert sind, sollte hier das Verhältnis 14 Marmoriert : 2 Einfarbig resultieren. Dieses Verhältnis konnte indessen nicht erhalten werden, es ergab sich 3 Marmoriert : 1 Einfarbig, so wie dies laut der EMERSON—SPILLMAN-schen (und auch PRAKKENS) Hypothese zu erwarten war. Wie aber die unten folgenden Kreuzungsergebnisse zeigen werden, bilden diese keineswegs eine Bestätigung dieser Hypothesen sondern lassen sie im Gegenteil unhaltbar erscheinen.

Kreuzung Nr. 144, Linie 127 × Linie 146. — Die Linie Nr. 127 wurde von mir aus einer aus Ungarn erhaltenen Samenprobe als Einmischung ausgelesen. Sie ist eine kleinsamige Wachsbohne. Die Samen sind schön Dunkel Weinrot marmoriert auf Rohseidengelbem Grund. Letzterer zeigt häufig eine schwache Rosa Tönung. Bei genauer Untersuchung sieht man deutlich, dass alle Samen ausser Marmorierung überdies die durch *S* bedingte Streifung aufweisen. An den Stellen, wo Marmorierung und Streifung sich decken, ist das Weinrot dunkler. Der zweite Elter, Linie 146, stammt aus meiner Kreuzung Nr. 12 (s. LAMPRECHT, 1932 a). L. 146 ist in allen Farbgenen rezessiv und hat nur die beiden Grundgene für Farbe, *P* und *Gri*, dominant. Da in Kreuzung Nr. 12, aus der sie herstammt, keine gestreiftsamigen Pflanzen aufgetreten sind, muss sie auch im Gen *S* rezessiv sein.

Die auf der ersten Generation erhaltenen Samen zeigten dasselbe Aussehen wie die des einen Elters, L. 127. In F_2 wurden die in Tab. 5 mitgeteilten Spaltungsergebnisse beobachtet.

Wie ersichtlich wurde für die rote Streifung nur das Symbol *S* benutzt. Dies geschah mit Hinblick auf die im vorigen Abschnitt mitgeteilten Resultate und Erörterungen, die es wahrscheinlich machten, dass *S* allein — also nicht *R* und *S* gemeinsam — für die Streifung ver-

antwortlich ist. Für die konstante Marmorierung wurde das dominante Gen *M* verwendet.

Die Kreuzung zeigte nur eine Zweigenenspaltung. Da auch in dieser F_2 mit 1033 Individuen keine Spaltung im Gen *R* stattfand, bildet dies eine weitere Bestätigung der eben erwähnten Annahme, dass *S* allein für die Ausbildung von Streifung und roter Farbe verantwortlich ist. Der eine Elter, L. 146, hatte ja bestimmt *R* in rezessiver Form. Ferner ist hier festzustellen, dass Marmorierung nur zusammen mit Streifung auftritt. Diese Erscheinungen dürften a priori in zweierlei Weisen erklärt werden können. 1) Das Gen *M* wirkt nur bei Dominanz in *S* (in Kreuzungen, die in *C* : *c* spalten, ist überdies zu berücksichtigen, dass *S* stark mit *C* gekoppelt ist); 2) *M* ist so stark mit *S* gekoppelt, dass auch bei der hier vorliegenden Individuenanzahl von 1033 kein Koppelungsbruch beobachtet werden kann.

TABELLE 5. *Die Aufspaltung des Bastards PP cc Jj gg bb vv Ss MM in F_2 von Kreuzung Nr. 144.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
F_2:	12 *J*	9 *SM.. ...*	Weinrot *SM/Rohsei-* dengelb	560	581,06	1,32
		3 *sM*	Rohseidengelb	213	193,69	1,54
	4 *j*	3 *SM*	Trübrosa *SM/Weiss*	179	193,69	1,17
		1 *sM*	Reinweiss..............	81	64,56	2,12

Es dürfte unschwer sein unter diesen Annahmen die naheliegendere zu wählen. Es soll dies ja stets auch die einfachste sein. Und das ist ganz zweifellos Hypothese Nr. 1. Für diese spricht auch ganz bestimmt, dass bisher keine konstant marmorierten Samen bekannt geworden sind, die nicht auch Streifung, *S*, gezeigt hätten. Die Hypothese, dass *M* nur bei Anwesenheit von *S* wirkt, erklärt auch völlig die vorhin angeführte Erscheinung, dass es bislang unmöglich gewesen ist, konstante Marmorierung in denselben Farben zu erhalten wie heterozygote, d. h. durch *Cc* bedingte. Denn im selben Augenblick, wo die Einführung von *S* in die Konstitution *Bedingung* dafür ist, dass überhaupt konstante Marmorierung erhalten werden kann, wird auch die Testafarbe durch den Roteffekt von *S* verändert. All dies spricht auch ganz entschieden gegen die Hypothesen von EMERSON—SPILLMAN und PRAKKEN, die konstante und heterozygote Marmorierung durch eine

gemeinsame genetische Grundlage erklären wollten. Weitere Belege
für die Unhaltbarkeit dieser Hypothesen werden Kreuzungen liefern, in
denen sowohl M wie C spalten.

Mit Hinblick auf Vorstehendes und in folgenden Kreuzungen fest-
gestellte Spaltungen in M bei gleichzeitiger Anwesenheit von S wurde
angenommen, dass L. 146 M in ihrer Konstitution hat. In bezug auf
die Bezeichnung der Testafarben sei hervorgehoben, dass die konstant
Marmorierten stets durch das Symbol SM nach der dunkleren, die
Flecken und Streifen bildenden Farbe angegeben werden, also z. B.
Weinrot SM/Rohseidengelb. Die Farben mit SM sind die gleichen wie
bei S allein (s. vorigen Abschnitt), nur dass die Farbe an den Stellen
der Testa, wo sich Marmorierung und Streifung decken, dunkler er-
scheint. Dies sei auch in die Bezeichnung SM einbegriffen. Die zweite,
nach dem / angeführte Farbe, gibt wie immer die des helleren Grun-
des an.

Kreuzung Nr. 149, Linie 9 × Linie 127. --- L. 9 siehe Kr. 58 in
vorigem Abschnitt. Sie hat die Testafarbe Amethystviolett S/Rohseiden-
gelb und die Formel $P\,J\,V\,S$. L. 127 s. vorige Kr. 144; ihre Testafarbe
ist Weinrot SM/Rohseidengelb, der Formel $P\,J\,S\,M$ entsprechend. Die
auf F_1 erhaltenen Samen stimmen in ihrer Färbung mit L. 9 überein,
nur waren sie marmoriert und gestreift. Ihre Farbenbezeichnung ist:
Amethystviolett SM/Rohseidengelb, $P\,J\,V\,S\,M$. In F_2 war eine Spaltung
in den zwei Genpaaren $V—v$ und $M—m$ zu erwarten. Dies traf zu.
Es wurden folgende Zahlen erhalten:

Gefunden: 299 $V\,M$: 121 $V\,m$: 89 $v\,M$: 27 $v\,m$
Erwartet: 301,5 » : 100,5 » : 100,5 » : 33,5 »
D/m für
9 : 3 : 3 : 1 — 0,22   + 2,27   — 1,27   — 1,16

Die erwarteten und gefundenen Zahlen zeigen befriedigende Überein-
stimmung. Als neu analysierte Farbe ergibt sich: *Amethystviolett SM/
Rohseidengelb, P J V S M.* Eine weitere Erläuterung zu dieser Kreu-
zung erscheint überflüssig, ihre Spaltung steht in voller Übereinstim-
mung mit den von mir vorhin gemachten Annahmen.

Kreuzung Nr. 312, Linie 124 × Linie 130. — L. 124 stammt aus der
bekannten deutschen Brechbohne Konserva mit bunten Samen. Diese
zeigen auf weissem Grund Trübrosa bis Lilarosa Marmorierung und
Streifung. Formel: $P\,S\,M$ = Trübrosa SM/Weiss. Linie 130 ist eine
Geschwisterlinie zu L. 118, die im vorigen Abschnitt in Kr. 142 beschrie-
ben worden ist. Sie hat die Formel: $P\,S$ = Trübrosa S/Weiss. F_1

zeigte die Testafarbe von L. 124. F_2 spaltete, wie zu erwarten war. nach 3 *SM* : 1 *Sm*. Es resultierte:

Gefunden: 420   Trübrosa *SM*/Weiss : 147   Trübrosa *S*/Weiss
Erwartet: 425,25   »   »   : 141,75   »   »
D/m für
    3 : 1 =   0,51.

Auch diese Spaltungsresultate bestätigen die vorhin angenommenen genetischen Grundlagen für Vererbung der konstanten Marmorierung.

TABELLE 6.   *Die Aufspaltung des Bastards PP cc Jj Gg bb vv Ss MM in F_2 von Kreuzung Nr. 197.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
F_2:	48 *J* { 36 *G* {	27 *SM*	Mahagonibraun *SM*/ Rohseidengelb ...	175	164,11	1,12
		9 *sM*	Maisgelb	65	54,70	1,52
	12 *g* {	9 *SM*	Weinrot *SM*/Rohseidengelb..............	56	54,70	0,19
		3 *sM*	Rohseidengelb	12	18,23	1,49
	16 *j* { 12 *G* {	9 *SM*	Hell Braunrot *SM*/ Speckweiss	51	54,70	0,54
		3 *sM*	Speckweiss	12	18,23	1,49
	4 *g* {	3 *SM*	Trübrosa *SM*/Weiss	13	18,23	1,25
		1 *sM*	Reinweiss	5	6,08	0,44

Diese wird stets durch das Zusammenwirken der beiden dominanten Gene *S* und *M* verursacht.

Kreuzung Nr. 197, Linie 42 × Linie 124. — Linie 42 stammt aus der bekannten deutschen Brechbohne Hundert für Eine. Ihre Samen sind einfarbig Maisgelb; Formel *P J G*. Linie 124 siehe vorige Kreuzung. Die auf F_1 erhaltenen Samen zeigten die Farbe Mahagonibraun *SM*/Rohseidengelb. F_2 zeigte die in Tab. 6 mitgeteilten Spaltungsresultate.

Die Spaltungsresultate von Kr. 197 bestätigen das früher Angeführte. Neu sind nur die Genanalysen der beiden Farben Mahagonibraun *SM*/Rohseidengelb und Hell Braunrot *SM*/Speckweiss. Auffallend ist, dass auch in dieser Kreuzung für den einfarbigen Elter, L. 42, angenommen werden muss, dass er *M* in seiner Konstitution hat. Das Gleiche war schon in Kr. 144 für Linie 146 der Fall.

Kreuzung Nr. 235, Linie 124 $\times$ Linie 161. — L. 124, Formel *P S M*, wurde schon vorhin erwähnt. L. 161 stammt aus der schwedischen Wachsbohne Express. Ihre Samen haben die Testafarbe Schamois, es ist ein lebhaftes Schamois. Formel *P C J* (möglicherweise *P C Ins*; ein sicherer Unterschied zwischen *Ins* und *J* ist noch nicht festgestellt). In dieser Kreuzung ist also eine gleichzeitige Spaltung in beiden Marmorierungen, konstanter und heterozygoter (*Cc*) zu erwarten. Die Samen der F_1-Generation zeigten in Übereinstimmung hiermit doppelte Marmorierung. Sie waren Pflaumenviolett *SM*/Schamois/Rohseidengelb.

TABELLE 7. *Die Aufspaltung des Bastards PP Cc Jj gg bb vv Ss MM in F_2 von Kreuzung Nr. 235.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
F_2:	4 *CC*	3 *J*	Schamois (hell-dunkler)......	94	101,44	0,82
		1 *j*	Geschwefeltes Weiss	35	33,81	0,21
	8 *Cc*	2 *J SSM*	Pflaumenviolett *SM*/Hell Schamois/Rohseidengelb...	55	67,62	1,84
		4 *J SM*..	Pflaumenviolett *SM*/Schamois/Rohseidengelb	145	135,25	0,97
		2 *j SM* ..	Hell Lila *SM*/Geschwefeltes Weiss/Weiss	66	67,62	0,21
	4 *cc*	1 *J SSM*	Weinrot *SM*/Hell Rohseidengelb......	36	33,81	0,39
		2 *J SM*..	Weinrot *SM*/Rohseidengelb	82	101,44	1,87
		1 *j SM*...	Trübrosa *SM*/Weiss	28	33,81	1,03

Die Tab. 7 zeigt, dass infolge der starken Koppelung zwischen *C* und *S* keine *CC*-Individuen mit *SM* erhalten wurden. Ferner ist ersichtlich, dass doppelte Marmorierung nur in der in *C* heterozygoten Individuengruppe aufgetreten ist. Die weitere Untersuchung der Kreuzung in F_3 ergab, dass diese doppelt marmorierten Pflanzen, wie zu erwarten, beständig spalteten, so wie dies übrigens für *Cc*-Marmorierung charakteristisch ist. Die Testafarben der doppelt marmorierten Samen ergeben auch, dass es sich bei der durch *SM* und der durch *Cc* bedingten Marmorierung um zwei genetisch getrennte Erscheinungen handelt. So setzt sich z. B. die Testafarbe Pflaumenviolett *SM*/Schamois/Rohseidengelb zusammen aus:

PP Cc JJ = Schamois/Rohseidengelb (heterozygot marmoriert) und
PP C J SM = Pflaumenviolett *SM*/Schamois (konstant marmoriert).

Aus der Tabelle ist ferner ersichtlich, dass die Individuen mit *J SM* in ihrer Konstitution in zwei Gruppen sortiert werden konnten, eine mit einer helleren und eine mit einer etwas dunkleren Farbe. Die für diese erhaltenen Zahlen stimmen gut auf das Verhältnis 1 : 2, d. h. Homo-: Heterozygotie in einem Gen. Es muss indessen einstweilen unentschieden verbleiben, ob dies mit Heterozygotie in *S* oder in *J* (vielleicht *Ins*) zusammenhängt. Hier sind spezielle Untersuchungen erforderlich.

Die vorstehend über die Vererbung von konstanter und beständig spaltender Marmorierung mitgeteilten Resultate zeigen zweifellos, dass diese beiden auf ganz verschiedener genetischer Grundlage stehen. Die beständig spaltende Marmorierung ist an Heterozygotie im Gen *C* gebunden, die konstante an das Gen *M* zusammen mit *S*. Durch *Cc* bedingte Testafarben können daher nicht durch *SM* erhalten werden und umgekehrt. Jede dieser Marmorierungen kann einzeln oder sie können auch gemeinsam auftreten. Im letzteren Fall erhält man doppelte Marmorierung, wobei die hellere Marmorierung durch *Cc* bedingt wird, die dunklere durch *SM*. Die von EMERSON—SPILLMAN und PRAKKEN aufgestellten, vorhin besprochenen Hypothesen sind mit diesen Tatsachen unvereinbar und daher als unhaltbar zu streichen.

Kann aber nun die von mir vorhin gemachte Annahme, dass *M* nur bei Dominanz in *S* wirkt, aufrecht erhalten werden? — Offenbar nur, wenn die von mir benutzten einfarbigen Linien alle *M* in ihrer Konstitution haben. Diese Annahme musste für den einen Elter in drei verschiedenen Kreuzungen gemacht werden, für L. 146 in Kr. 144, für L. 42 in Kr. 197 und für L. 161 in Kr. 235. Diese Annahme erschien wenig wahrscheinlich und führte den Gedanken darauf, dass es sich bei den Genen *S* und *M* um multiple Allele von *R* handeln könnte. Zur Bestätigung dieser Annahme ausgeführter Kreuzungen ergaben die erwarteten Resultate. Eine dieser, Nr. 111, ausgeführt zwischen L. 9 und L. 146 (beide vorstehend beschriebene Linien) genügte um drei Allelen dieser Serie festzulegen. Sie spaltete in F_2 in 227 gestreift : 81 einfarbig. D/m ist hierfür 0,53. Es ergeben sich dann zusammen mit den zwei oben besprochenen Kreuzungen Nr. 144 und Nr. 149 folgende Spaltungen:

Kreuzung Nr. 144: L. 146 × L. 127: 3 konstant marmoriert : 1 einfarbig;
 »   .Nr. 149: L. 127 × L. 9: 3 » » : 1 gestreift;
 »   Nr. 111: L. 9 × L. 146: 3 gestreift : 1 einfarbig.

Durch dieses Kreuzungsdreieck ist *die multiple Allelie von M und S mit r in der R-Serie bewiesen.* Über die Zugehörigkeit von *R*, des vierten Allels zur Serie, kann kein Zweifel mehr bestehen, da Kreuzungen von *R*- mit *r*-Linien (worunter sich auch oben genannte befinden) stets monohybrid 3 *R* : 1 *r* gespalten haben.

In Übereinstimmung mit diesen Ergebnissen sind die beiden Gene *S* und *M* zu löschen und als multiple Allele an *R* anzugliedern. Für diese Allelen werden folgende Bezeichnungen vorgeschlagen:

R_{ma} = Trübrosa marmoriert und gestreift auf Weiss = Trübrosa R_{ma}/Weiss,

R_{st} = Trübrosa gestreift auf Weiss = Trübrosa R_{st}/Weiss,

R = Trübrosa einfarbig, und

r = Reinweiss.

Diese Testafarben gelten natürlich nur bei Dominanz in den Grundgenen für Testafarbe *P* und *Gri* sowie bei Rezessivität in allen anderen Farbgenen. Bei Dominanz in letzteren werden verschiedene Farben in marmoriert, gestreift, bzw. einfarbig erhalten. *R* (und seine Allelen) sind stark mit *C* gekoppelt. Die Dominanzreihenfolge der vier Allelen ist noch nicht vollkommen klargelegt.

Im folgenden seien die bisher genetisch analysierten konstant marmorierten Testafarben angeführt:

Trübrosa R_{ma}/Weiss, *P c j g b v* R_{ma}

Weinrot R_{ma}/Rohseidengelb, *P c J g b v* R_{ma}

Hell Braunrot R_{ma}/Speckweiss, *P c j G b v* R_{ma}

Mahagonibraun R_{ma}/Rohseidengelb, *P c J G b v* R_{ma}

Amethystviolett R_{ma}/Rohseidengelb, *P c J g b V* R_{ma}

An doppelt marmorierten wurden analysiert:

Pflaumenviolett R_{ma}/Schamois/Rohseidengelb, *P Cc J g b v* R_{ma}

Hell Lila R_{ma}/Geschwefeltes Weiss/Weiss, *P Cc j g b v* R_{ma}

Sämtliche vorstehend besprochenen Kreuzungen wurden auch in F_3 untersucht und stimmten die Resultate mit den auf Grund der F_2-Spaltung erwarteten überein.

DIE VERERBUNG DER BESPRITZUNG DER TESTA.

In einer früheren Arbeit (LAMPRECHT, 1934 b, S. 182) wurde der sog. gespritzte Typus von *Phaseolus vulgaris* beschrieben. Ich erhielt diesen seinerzeit u. a. unter der Bezeichnung *Ph. atropunctatus* aus

Botanischen Gärten. Es handelte sich aber um keine selbständige Art sondern nur um eine Varietät der gewöhnlichen Gartenbohne. Diese Form gab bei Kreuzung mit *Phaseolus vulgaris*-Linien durchweg voll fertile Nachkommen und zeigte i. ü. keine abweichenden Charaktere. Ihrem Habitus nach gehört sie zu den sog. Reiserbohnen. Diese haben bekanntlich unbegrenztes Stammwachstum, aber kurze Internodien, sodass sie eine Höhe von 60—70 cm erreichen. Eine Linie vom gespritzten Typus, L. 53, wurde zu einer Anzahl von Kreuzungen verwendet. Fig. 5 zeigt die Zeichnung der Samen dieser. Wie ersichtlich ist die Testa auf hellerem Grund gleichsam mit feinen, unregelmässigen Pünktchen reichlich bespritzt. Eine Vergrösserung dieser Zeichnung zeigt Fig. 6. Ausserdem gewahrt man unregelmässig verteilte grössere, dunkle Farbflecken. Die Farbe dieser sowie der Pünktchen ist Schwarz
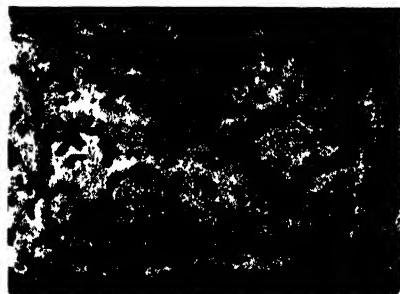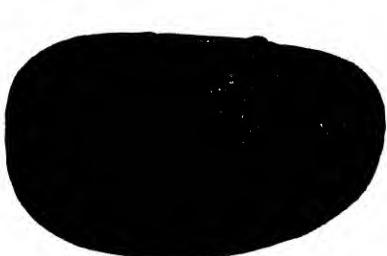
Fig. 5. Ein Samen mit Bespritzung der Testa aus Linie 53 (sog. *Phaseolus atropunctatus*). — Fig. 6. Die Zeichnung der Testa eines bespritzten Samens bei etwa 60-facher Vergrösserung.

bis Violettschwarz. An gewissen Samen kann man deutlich erkennen, dass diese grösseren Flecken Rudimente der durch das Allel R_{st} bedingten Streifen darstellen (vgl. die vorigen Abschnitte). Eine genetische Analyse dieser Bespritzung der Testa scheint bisher nicht erfolgt zu sein.

Kreuzung Nr. 171, Linie 1 × Linie 53. — L. 1 stammt aus der schwedischen braunen Kochbohne Stella. Diese wurde mehrmals genetisch untersucht. Hinsichtlich Testafarbe kommt ihr die Konstitution *P C J G b v r* zu. Über die Formel von L. 53, mit Violettschwarz bespritzter Testa war bisher nichts bekannt. Die auf der ersten Generation erhaltenen Samen zeigten die Testafarbe Schwarz *Cc*-marmoriert/Graulich Indigo mit Violettschwarzer Bespritzung des Grundes. Die Grundfarbe Graulich Indigo ist selten gut ausgebildet sondern meistens Graulich Rhamninbraun mit mehr oder weniger deutlichem Graulich Indigo Anflug. Der Testafarbe Schwarz/Graulich Indigo kommt

laut früheren Analysen folgende Formel zu: $P\ Cc\ J\ G\ B\ V$; im vorliegenden Fall wäre das neue Gen für die Bespritzung hinzuzufügen. Da die auf F_1 erhaltenen Samen bespritzte Testa zeigen, hat man es mit einer dominanten Eigenschaft zu tun. Da die Samen marmoriert sind,

TABELLE 8.   *Die Aufspaltung des Bastards* $PP\ Cc\ JJ\ GG\ Bb\ Vv\ R_{res}r$
in F_2 *von Kreuzung Nr. 171.*

Genenspaltung			Testafarben	Anzahl Individuen		D/m
				Gefunden	Erwartet	
$F_2{:}JG$	16 CC	12 B { 9 $V\,r$ ...	Schwarz	107	113,48	0,66
		3 $v\,r$ ...	Mineralbraun..............	38	37,83	0,03
		4 b { 3 $V\,r$ ...	Kastanienbraun..........	28	37,83	1,63
		1 $v\,r$ ...	Bister.....................	14	12,61	0,40
	32 Cc	24 B { 18 $V\,R_{res}$	Violettschwarz R_{res}/ /Schwarz'Graulich Indigo	247	226,97	1,57
		6 $v\,R_{res}$	Purpurschwarz R_{res}/Mineralbraun/Rhamninbraun......................	91	75,66	1,86
		8 b { 6 $V\,R_{res}$	Dkl. Purpur R_{res}/Kastanienbraun/Ageratumblau	63	75,66	1,53
		2 $v\,R_{res}$	Vandyke Rot R_{res}/Bister/ /Maisgelb	17	25,32	1,68
	16 cc	12 B { 9 $V\,R_{res}$	Violettschwarz R_{res}/ /Graulich Indigo	122	113,48	0,88
		3 $v\,R_{res}$	Purpurschwarz R_{res}/ /Rhamninbraun.........	32	37,83	0,97
		4 b { 3 $V\,R_{res}$	Dkl. Purpur R_{res}/Ageratumblau	29	37,83	1,47
		1 $v\,R_{res}$	Vandyke Rot R_{res}/Maisgelb	19	12,61	1,81

soll L. 53 c in ihrer Konstitution haben, denn L. 1 hat G und der heterozygoten Marmorierung von F_1 entspricht Cc. Die in F_2 beobachteten Spaltungsresultate sind in Tab. 8 zusammengestellt.

Tab. 8 zeigt, dass wir es hier mit einer Spaltung in 4 Genen zu tun haben, und zwar in den gut bekannten Testafarbgenen C, B und V sowie in dem Gen, das die Bespritzung der Testa bedingt. Auf Grund der

erhaltenen Farben ist ferner ersichtlich, dass beide Eltern in den Genen J und G dominant sein müssen. Es wurden 8 verschiedene homozygote Farben und dementsprechend 4 in C heterozygote, marmorierte gefunden. Von besonderem Interesse ist nun die Erscheinung, dass die Bespritzung der Testa nur auf Cc- und cc-Samen auftritt. Da auch die Elternlinie 53 mit bespritzter Testa cc in ihrer Konstitution hatte, spricht dies für eine sehr starke bzw. absolute Koppelung zwischen C und dem noch unbekannten Gen für Bespritzung der Testa. Ferner ergibt sich aus den Testafarben der bespritzten Typen, dass diese ein Gen für Rotfärbung enthalten müssen. Als solches ist bisher nur R bekannt. Laut der früheren Auffassung von roter Streifung und Marmorierung, als durch R, S bzw. M bedingt, wäre hier abermals ein neues Gen anzunehmen, das zusammen mit R rote Bespritzung der Testa verursacht. Und dann würde man hinsichtlich der Beziehung dieses neuen Gens zu R in genau die gleiche Lage gelangen wie sie früher für S und M vorhanden war, d. h. es wäre absolute Koppelung auch zwischen dem neuen Gen und R anzunehmen. Wenn hierzu noch die starke Koppelung mit dem Testafarbgen C kommt, die auch für die R-Allelenserie gilt, und schliesslich festgestellt werden kann, dass die Testa der bespritzten Samen überdies deutliche Reste der durch das Allel R_{st} (früher S) bedingten Streifen aufweist, dürfte meiner Ansicht kein Zweifel darüber zu bestehen brauchen, dass wir es hier mit einem neuen, fünften Allel der R-Serie zu tun haben. Dieses neue Allel will ich mit dem Symbol R_{res} bezeichnen, abgeleitet von *respergere* = bespritzen. Die Ordnungsfolge der nun bekannten fünf Allelen von R ist, wie schon aus dem vorigen Abschnitt hervorgeht, nur zum Teil sichergestellt.

In der vorliegenden Kreuzung Nr. 171 wurden folgende vier Typen mit homozygoter bespritzter Testa genetisch analysiert:

Violettschwarz R_{res}/Graulich Indigo: $P\ c\ J\ G\ B\ V\ R_{res}$

Purpurschwarz R_{res}/Rhamninbraun: $P\ c\ J\ G\ B\ v\ R_{res}$; die Farbe der Bespritzung entspricht CS, XLIV, 65''' m.

Dkl. Purpur R_{res}/Ageratumblau: $P\ c\ J\ G\ b\ V\ R_{res}$; Farbe der Bespritzung = CS, XII, 67 m. Die Grundfarbe Ageratumblau ist selten typisch sondern gewöhnlich Zimmtbraun mit mehr oder weniger deutlichem Anflug von Ageratumblau. In gleicher Weise ist die Grundfarbe Graulich Indigo häufig Rhamninbraun mit Graulich Indigo Anflug.

Vandyke Rot R_{res}/Maisgelb: $P\ c\ J\ G\ b\ v\ R_{res}$; Farbe der Bespritzung = CS, XIII, 1' k.

ÜBER DIE FÜNF ALLELEN DES GENS R.

In den drei vorstehenden Abschnitten wurden Vererbungsstudien über Streifung, konstante Marmorierung und Bespritzung der Testa von *Ph. vulgaris* mitgeteilt, die schliesslich zu der Erkenntnis führten, dass die für diese Eigenschaften verantwortlichen Gene zu einer Serie von fünf Allelen des Gens *R* gehören. Eine kurze zusammenfassende Betrachtung über diese mag hier am Platze erscheinen.

Die Vererbung von roten, rötlichen bis dunkelvioletten Testafarben ist von Genetikern relativ wenig studiert worden. SHAW and NORTON (1918) teilten die Testafarben von *Ph. vulgaris* in zwei Serien ein, eine Gelb-Schwarz- und eine Rot-Serie, für die als solche je ein Gen *M* bzw. *M'* (eine Art Grundgen) verantwortlich gemacht wurde. Kreuzungsergebnisse, die diese Annahme bestätigten, wurden aber nicht beigebracht. TJEBBES (1931) schreibt die Ausbildung von roten Testafarben einem Farbgen *R* zu. LAMPRECHT (1935) studierte die Wirkung dieses Gens zusammen mit den vorher gut analysierten Farbgenen *C* und *J* und zeigte u. a., dass *R* in heterozygoter Form mehr oder weniger deutlich ausgebildete Marmorierung der Testa bedingt.

Das Gen *R* hat gleich den anderen Farbgenen von *Ph. vulgaris*: *C, J, Ins, Can, G, B* und *V*, zusammen mit den Grundgenen für Testafarbe *P* und *Gri* in dominanter Form eine bestimmte Farbenwirkung auf die Testa. *P Gri R*-Samen sind Trübrosa gefärbt. *P Gri r*-Samen sind Reinweiss. Zusammen mit den anderen Farbgenen gibt *R* verschiedene rötliche-braunrote-violette-schwarze Farben der Testa. — Seit langem war nun auch bekannt, dass die Farbe Trübrosa auch gestreift (Fig. 2) und konstant marmoriert (Fig. 3) auf Reinweiss vorkommt (die Sorten Heinrichs Riesen und Konserva mit bunten Bohnen). Auch andere rötliche, braunrote und dunklere violett-blauschwarze-schwarze Streifung und Marmorierung kommt vor. Für die Ausbildung von Streifung wurde ein besonderes Gen *S* (E. v. TSCHERMAK, 1912) und für Marmorierung *M* (SHULL, 1908) angenommen. Spätere Untersuchungen von TJEBBES und KOOIMAN (1921 a und 1921 b) zeigten, dass *R* und *S* ausserordentlich stark (absolut?) gekoppelt sind. Über das Gen *M* für konstante Marmorierung lagen bisher keine diesbezüglich klarlegenden Studien vor. Einige Forscher unternahmen den Versuch die durch Heterozygotie in *C* bedingte und die konstante Marmorierung, *M*, durch Hypothesen genetisch unter einen Hut zu bringen (EMERSON, 1909 b; E. v. TSCHERMAK, 1912; PRAKKEN, 1934).

Auffallend war nun, dass Streifung und Marmorierung stets nur

in gewissen Farben angetroffen wurden, d. h. in solchen, die durch das Gen R, allein oder in verschiedenen Kombinationen mit anderen Farbgenen, bedingt werden. Die vorliegende Arbeit zeigte nun, dass die Gene S, M und r bzw. R verschiedene Allelen desselben Gens darstellen. Dadurch war natürlich auch die Unmöglichkeit bewiesen, zwischen den vermeintlich selbständigen, d. h. von R unabhängigen, Genen S bzw. M und R einen Koppelungsbruch anzutreffen.

Die vorstehend mitgeteilten Beobachtungen zwingen also dazu, die früher für Streifung und konstante Marmorierung der Testa benutzten Gensymbole S und M zu streichen und durch Allele zu R zu ersetzen. Als solche wurden R_{st} und R_{ma} gewählt. Die von TJEBBES (1931) aufgestellte Koppelungsgruppe C (von ihm mit dem schon früher vergebenen Symbol B bezeichnet) —R—S wird hierdurch auf nur zwei Gene, C und R, reduziert. In vorliegender Arbeit wurde schliesslich eine neue Zeichnung der Testa, sog. Bespritzung, genetisch analysiert. Auch diese wird durch ein Rotgen bedingt und es konnte gezeigt werden, dass das betreffende Gen sich genau so verhält wie früher gerade für S und M angenommen worden ist. Dieses neue Gen stellt mit grösster Wahrscheinlichkeit ein weiteres Allel der R-Serie dar und wurde in Übereinstimmung hiermit mit dem Symbol R_{res} belegt.

Die vorgelegten Resultate sind auch entscheidend für die Haltbarkeit der von EMERSON, v. TSCHERMAK und PRAKKEN aufgestellten Hypothesen zur Erklärung der Vererbung von heterozygoter und konstanter Marmorierung. Diese sind als unhaltbar zu streichen. Eine nähere Erörterung hierüber findet sich in dem Abschnitt über Marmorierung.

Hervorgehoben zu werden verdient schliesslich die Erscheinung, dass der marmorierte Typus auch gleichzeitig Streifung aufweist. Die Wirkung des Allels R_{ma}, das ja über R_{st} dominiert (Spaltung 3 R_{ma} : 1 R_{st}), löscht die Wirkung des letzteren Allels nicht aus, oder besser gesagt R_{ma} bedingt also sowohl Marmorierung wie Streifung der Testa. Bei dunkleren Farben ist dies allerdings schwer zu erkennen. Ähnliches kann auch in gewissem Grade von R_{res} behauptet werden, denn dieses Allel bedingt auf der Testa nicht nur Bespritzung mit feinen, unregelmässigen Punkten sondern die Testa zeigt überdies grössere Fleckchen in der Farbe der Bespritzung, die oftmals deutlich als Rudimente der durch R_{st} bedingten Streifung erkannt werden können. Die feineren Fleckchen und Pünktchen der Bespritzung könnten dann die durch die Wirkung des Allels R_{res} aufgelöste Marmorierung darstellen. Ausgehend von dieser Überlegung würde man dann als wahrscheinliche

Reihenfolge für die Allelen von R folgende von Dominanz nach Rezessivität erhalten: R_{res}—R_{ma}—R_{st}—R—r.

SUMMARY.

1. The inheritance of the different types of the polychromatic seed coat of *Phaseolus vulgaris* is studied.

2. These types are the following: a) striped, b) homozygous marbled, a type which moreover always is striped, c) heterozygous marbled, and d) sprinkled.

3. It could be stated that striped, homozygous (constant) marbled and sprinkled seed coat only appears, when the gene R (for light roseate seed coat colour) is dominant. In connection with this phenomenon the genes, hitherto used for striping, S, and constant marbling, M, seem to be absolutely linked with R.

4. Crosses between three pure lines with the constitution $R\,s\,m$, $R\,S\,m$ and $R\,s\,M$ showed all monohybrid segregation 3 : 1. Herewith it was proved that the three genes R, S and M are different allels of the same gene R.

5. R shows strong linkage with the seed coat colour gene C (already stated by TJEBBES). The same is naturally the case with S and M.

6. In the present paper a new gene is stated for sprinkled seed coat. This gene is exactly in the same way as S and M absolutely linked with R and shows strong linkage with C. It is evident a new allel of the R-series.

7. The hitherto known allels of the R-series are signified in the following way: R_{res}—R_{ma}—R_{st}—R—r.

8. The heterozygous marbling of the seed coat is caused by heterozygosity in C respectively in R. The Cc-marbling is always strong, the Rr-marbling contrary more or less indistinct.

9. In connection with the fact that constant marbling is caused by the allel R_{ma} (which moreover causes red colour), the Cc-marbling never occurs in the same colours (excepting black).

10. The hypotheses of EMERSON—SPILLMAN, E. V. TSCHERMAK and PRAKKEN to explain the inheritance of heterozygous and homozygous marbling from the same genetical point of view must now be considered as untenable.

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INHALT.

FURTHER CONTRIBUTIONS TO A CHROMOMERE ANALYSIS OF LILIUM

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IN a recent paper (HEILBORN, 1939), the writer presented preliminary data on the chromomere structure of the pachytene chromosomes in *Lilium umbellatum*. The chromomeres were described as chromatic discs separated by achromatic material, the pachytene chromosomes presenting an appearance essentially the same as that of the salivary gland chromosomes of Diptera. This discovery being of a certain general importance, further research seemed necessary. The main purpose of such a continued research may be summed up as follows: to gather more complete data regarding the chromomeres in *Lilium*; to investigate the possibilities of mapping the pachytene chromosomes with regard to their chromomeres; to investigate other plants, too, thus examining the possibilities of establishing a chromomere analysis of a more general scope.

As the work proceeded, the writer soon realized that very great difficulties are met with. Positive results have as yet been obtained only with two species of *Lilium* (*L. umbellatum* and *L. longiflorum*) and the most evident new contribution is presented by a series of micro-photographs which are considered to give a good idea of the chromomeres of these species. As regards the mapping of the chromosomes and the investigation of other plants, but little progress has been made. Thus before proceeding to a more detailed description of the chromomeres in *Lilium*, a discussion of the *technical* and *biological* conditions for a chromomere analysis of plants seems to be appropriate.

TECHNICAL AND BIOLOGICAL CONDITIONS FOR A CHROMOMERE ANALYSIS OF PLANTS.

The technique used in the present investigation has been described in earlier papers (HEILBORN, 1937, 1939). As already pointed out (l. c. 1939, p. 106), the cell walls of the pollen mother-cells present a special difficulty. In order to spread and stretch the chromosomes — thus making the chromomeres stand out clearly — it is necessary to

squeeze the pollen mother-cells in such a way as to cause *the nuclei to escape out of the cell walls.* At the same time, the nuclear membrane, too, must be crushed and the chromosomes spread. When a piece of tissue from the interior of an anther is smeared on a slide, the majority of the pollen mother-cells are only crushed and flattened by the pressure on the cover glass, and the nuclei do not emerge. However, round the edge of the smear a border of plasm (without cell walls) appears and a number of nuclei are pushed into this plasm. A small proportion of these latter nuclei may have their chromosomes favourably spread. Plate I, Fig. 1, shows a piece of such a smear with flattened cell walls to the left and at the top, and plasm with nuclei to the right and at the bottom.

The pressure on the cover glass has this effect only when the amount of tissue smeared is not too small. If it is *very* small, the method is apt to fail (the nuclei then do not escape out of the flattened pollen mother-cells). Consequently, the method works relatively well only with plants whose anthers have attained a considerable size already at the time when their pollen mother-cells enter the pachytene stage. A large size is required also for dissecting the anthers and removing the anther walls.

Another technical difficulty lies in the extreme delicacy of the chromomeres. They are easily damaged. — Moreover, a successful chromomere analysis requires plant material with few and large chromosomes.

Obstacles of a purely biological kind are also met with. The writer has observed repeatedly that fully developed chromomeres can be obtained only *at the very end of the pachytene stage,* immediately before the four chromatids separate at the beginning of diplotene. The general conditions for a good development of the chromomeres are obviously the same in the anthers of plants as in the salivary glands of the larvae of Diptera. *In both cases, a prolonged prophase appears to be necessary.* The termination of this prophase will be the favourable moment for the investigator. A little later, the salivary gland tissue breaks down at the pupation of the larvae, the chromatids separate at the beginning of diplotene in the pollen mother-cells. The transitional stages between the fully developed pachytene and diplotene pass rapidly. Hence, the appropriate stage is of short duration, at least in *Lilium,* and is difficult to catch.

Low temperatures have a favourable effect upon the development of the chromomere discs in the salivary glands. The same thing may

be assumed with regard to the pachytene chromomeres. Plant ex-
periments with temperatures slightly above zero seem, therefore, very
desirable.

Consequently, for a successful chromomere analysis of plants the
fulfilment of the following conditions seems to be necessary:

The plants should have large anthers and at the same time few
and large chromosomes. A relatively large amount of pollen mother-
cell tissue should be used when preparing the smears. The chromo-
somes should be smeared at the very end of the pachytene stage. This
stage is short.

THE CHROMOMERES IN LILIUM.

General. — When the pachytene reaches its terminal stage, the
chromomeres appear as sharply outlined discs, some thin, some thick.
The conjugation of the chromosomes is complete, and the chromomeres
are seemingly quite homogeneous and uniform. All the chromosomes
in Text-fig. 1 and Plate I belong to this stage. Somewhat later, when
the chromatids separate at the beginning of diplotene, the discs become
quadripartite, each consisting of four small chromomeres. The internal
structure of the chromosomes now becomes somewhat indistinct, and
the nuclei are no longer quite suitable for cytogenetic purposes.

As pointed out in my earlier paper (1. c. 1939), some parts of the
chromosomes contain preponderatingly large chromomeres, others
smaller ones, in still other parts there is a mixture of different size-
classes. The largest chromomeres appear as »knobs». The chromo-
some arm to the left in Pl. I, Fig. 2, contains several thick chromomeres,
among them one »knob», near the middle of the same figure is an arm
with numerous, slender chromomeres. To the left of Fig. 4 (Pl. I) is
another arm with mostly slender chromomeres. Figs. 5—6 (Pl. I)
show various size-classes. The chromosome end of Fig. 7 (Pl. I)
contains one »knob» and, in addition, five slender discs.

In my previous paper (1. c. 1939, p. 107) a difference between
Lilium and *Drosophila* was touched upon: the »knobs» of *Lilium* are
never seen in *Drosophila*, and the numerous, exceedingly delicate chro-
momeres of *Drosophila* seemed to be absent in *Lilium*. The last-
mentioned statement is confirmed by the study of the writer's new
material. Hence, though the chromosome structure is essentially the
same in both types of organisms, the general appearance is somewhat

different. This circumstance seems to open up a possibility for a successful *comparative* analysis of chromomeres.

Pairs of chromomeres. — As pointed out earlier (l. c. 1939), at many points in the chromosomes discs of equal size appear to lie in pairs. At certain points as many as 3—5 such equally-sized discs may be found together. When a chromosome is properly stretched, such discs of equal size may be seen lying close together but separated from

Fig 1 Parts of pachytene chromosomes in *Lilium*, showing chromomeres — *a—d L umbellatum e L longiflorum* — *a* Chromomeres of equal size lying in pairs (cf Pl I, Fig 6) — *b* End of a chromosome with one »knob» and five slender discs, the two outermost of equal size, constituting a pair (cf Pl I, Fig 7) — *c* Another chromosome end (cf Pl I, Fig 8) — *d* A third chromosome end — *e* Chromomeres of equal size lying in groups of 3—5, groups separated by wide achromatic gaps (cf Pl I, Fig 2)

adjacent chromomeres or groups of chromomeres by somewhat wider achromatic pieces· Pl. I, Fig. 2 (cf. Text-fig. 1 *e*) and Fig. 6 (cf. Text-fig. 1 *a*). One further case of double chromomeres is seen in Pl. I, Fig. 5, and another at the very tip of the chromosome end in Pl. I, Fig. 7 (cf. Text-fig. 1 *b*). The *frequency* of paired chromomeres seems to be about the same in both *Lilium* species investigated. In some fairly clear chromosome pieces analyzed, out of 112 chromomeres counted in *L. umbellatum*, 35 or 31 % were found in pairs (or in groups

of 3—5); in *L. longiflorum* 32 chromomeres of 85 or 38 % were likewise in pairs or groups.

Paired chromomeres have probably arisen by duplication of originally single ones. Such duplicated chromomeres are a characteristic feature of many parts of the salivary gland chromosomes of *Drosophila* (e. g. the innermost part of the left arm of the third chromosome of *D. melanogaster*). Similarly, in *Sciara* METZ (1938) reports numerous clear cases of small duplications of discs. Unequal crossing-over may be conceived as a possible mode of origin (cf. the very instructive discussion by METZ, p. 284). However, nothing particular is known about this point, nor do we know much about the genetic effects (though some such effects may be taken for granted).

The first result of a chromomere duplication is a pair of chromomeres. Repeated duplications at the same locus will then give rise to rows of several equally-sized discs, and sometimes the process will result in the condition, outlined above, in which some parts of the chromosomes contain large chromomeres only, others slender ones. We have here two different observations, made independently, one of characteristic differences in the distribution of thick and slender chromomeres, and another of pairs of chromomeres of equal size, but both are probably only two phases of the same phenomenon.

Repeated duplications of chromomeres may be supposed to have given rise to a gradual increase in the length of the chromosomes. In fact, the chromosomes of *Lilium* are known to be unusually long! On the whole, the long chromosomes of most higher organisms may have been gradually built up of chromomeres in some such way. This interpretation is speculative, it is true, but it seems to have a certain resemblance to the theoretical views of some authors concerning supposed phylogenetic changes of chromosomes through »gradual growth» (cf. REUTER's discussion, 1930, pp. 128, 141).

Chromosome ends. — It is very difficult (practically impossible) to arrive at a complete chromomere map of the nucleus of a *Lilium* species. The same thing must be true of a great many other organisms. However. for certain cytogenetic purposes a knowledge of the structure of the chromosome *ends* only would suffice to furnish much valuable information. Thus, for instance, the cytogenetics of *Datura* is almost exclusively founded upon the ends of the chromosomes. Likewise, in the cytogenetics of *Oenothera* and other cases of segmental interchange the chromosome ends play an important rôle.

I have tried to examine the possibilities of such a type of chro-

momere analysis by searching for well-prepared ends of the chromosomes of *Lilium umbellatum*. Unfortunately, the result is very meagre. Chromomere maps of three such ends (out of 24 ends, corresponding to 12 haploid chromosomes) are presented in Text-fig. 1 *b—d* (cf. also Pl. I, Figs. 7—8). As seen, the differences in structure are considerable. It is true, that I do not as yet know anything about the *constancy* of these structural differences, but in view of all experience from *Drosophila* and *Zea mays*, as well as, recently, also *Antirrhinum* (ERNST, 1939) such a constancy need not be doubted.

SUMMARY.

The work presents new observations on the chromomeres of two species of *Lilium*, in addition to the preliminary data in the writer's previous paper on the same subject. The technical possibilities of an analysis of the chromomeres are discussed. An important biological circumstance to be considered is the prolonged prophase that seems to be required for a good development of chromomeres. This conclusion is based on the observation that well-developed chromomeres are seen only at the very end of the pachytene stage, immediately before the chromatids separate at the beginning of diplotene. It is also in good agreement with the well-known fact that the chromomeres in the salivary glands of Diptera do not attain their full size until just before the pupation of the larvae. At the end of the pachytene of *Lilium* the chromomeres have the shape of sharply outlined discs, separated by nonchromatic parts of the chromosomes. These discs look quite homogeneous: that each of them is composed of four small chromomeres, is not shown until the beginning of diplotene. Chromomeres of equal size often lie in pairs, sometimes in groups of 3—5. This phenomenon is regarded as being due to duplications of single chromomeres. Such processes have probably caused a characteristic distribution of the chromomeres, often occurring and observed already earlier, viz. that some parts of the chromosomes contain mostly thick chromomeres, and others slender ones. It may also be concluded that repeated duplications of chromomeres result in a gradual increase in chromosome length, a circumstance of a certain phylogenetic interest. A difference between *Lilium* and *Drosophila* is recorded: the former genus lacks the many very fine discs characteristic of the latter, but has, instead, a number of big »knobs» which are not found in *Drosophila*. Chromomere maps

of a few ends of chromosomes are presented as a tentative contribution to a more complete cytologic mapping.

Uppsala, October 1939.

EXPLANATION OF PLATE I.

Microphotographs of smeared pollen mother-cells and pachytene chromosomes of *Lilium*. Fig. 1: 100 ✕. Fig. 3: 1300 ✕. Figs. 2, 4, 7 and 8: 1800 ✕. Fig. 6: 1900 ✕. Fig. 5: 2300 ✕. Figs. 1—5: *Lilium longiflorum*. Figs. 6—8: *Lilium umbellatum*. — Fig. 1: Smeared pollen mother-cell tissue with flattened mother-cells and empty cell walls to the left and at the top, plasm and nuclei, squeezed out of the cells, to the right and at the bottom (cf. text). — Figs. 2—4: Pachytene chromosomes with discoid chromomeres, separated by non-chromatic material (Fig. 2: cf. Text-fig. 1 *e*). — Fig. 5: Detail of Fig. 3. — Fig. 6: Chromomeres *in pairs* (lower chromosome arm; cf. Text-fig. 1 *a*). — Figs. 7—8: Two chromosome *ends*, showing different chromomere structure (cf. Text-fig. 1 *b—c*).

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TRIPLOIDY IN TRITON TAENIATUS LAUR.

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(Preliminary Note)

RECENT investigations would appear to show that polyploidy, especially triploidy, is not too rare among amphibians. Triploid individuals have been found among *Anura* within the species *Rana esculenta* (G. and P. HERTWIG, 1920; DALCQ, 1930), *Rana fusca* (DALCQ, 1930) and *Rana pipiens* (PARMENTER, 1933) as well as among *Urodela* within the species *Triton palmatus* (FANKHAUSER, 1934) and *Triturus viridescens* (FANKHAUSER and KAYLOR, 1935). In all of these cases the findings were made on larvae or embryos from preserved material. More recently, FANKHAUSER (1938 and 1939) demonstrated the occurrence of triploidy in natural populations of *Triturus viridescens* and *Eurycea bislineata*; among the latter tetraploids were also discovered. In these two species the number of chromosomes was determined by studies of mitoses in the tail epithelium and connective tissue of the larvae, tissues of different embryogenic origin. In *Triturus viridescens* the development of the larvae up to metamorphosis was studied by FANKHAUSER.

In the course of an investigation on the spermatogenesis in *Triton tœniatus* LAUR. (syn. *Triturus vulgaris* L.), a species very common in Sweden, an individual with triploid testes was found. As triploidy was not the object of investigation at the time, the other parts of the animal were unfortunately thrown away, and hence it is not possible to assert definitely that all the cells were triploid.

Not much can be said, of course, about the individual from which these triploid testes were taken and fixed (in BOUIN-ALLEN's fluid). In general habitus it did not present any deviations from normal diploids. Its size was certainly above average, though quite within the range of variation. This agrees with FANKHAUSER's finding (1939 a) in *Triturus viridescens*.

Histology of the testes. — A cytological study of the meiosis in this triploid is in progress. Abundant divisions of spermatocytes and pre-spermatids were observed. The first metaphase shows the chromosome configurations well known from the meiotic divisions in triploid plants

(cf. Figs. 1 and 13). It would appear to be the first time these configurations have been observed in a triploid animal within the vertebrate phylum. Fig. 1 shows a completely analysed first metaphase in the
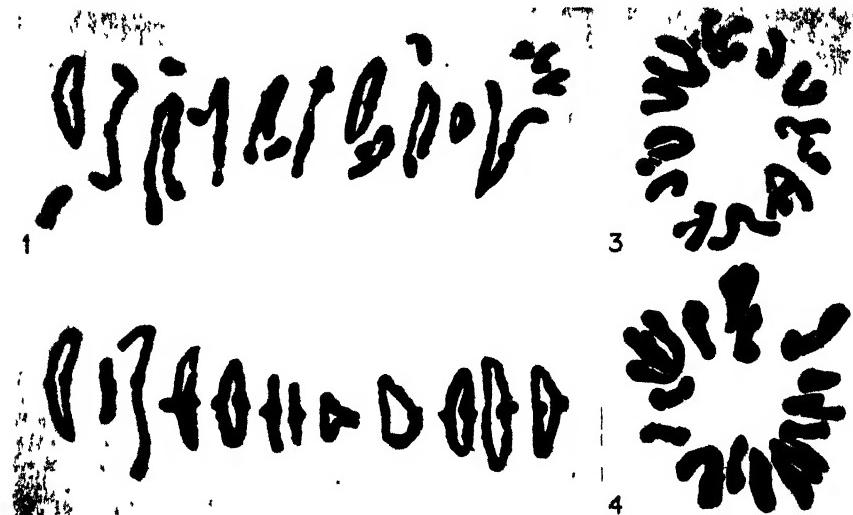

Fig. 1. First metaphase chromosomes of spermiogenesis in triploid testis. 2n = 36. (× 1340). — Fig. 2. First metaphase bivalents of spermiogenesis in diploid. 2n = 24. (× 1340). — Fig. 3. Second metaphase of spermiogenesis in diploid. 12 chromosomes. (× 2100). — Fig. 4. Second metaphase of spermiogenesis in triploid testis. In this instance 15 chromosomes. (× 2300).
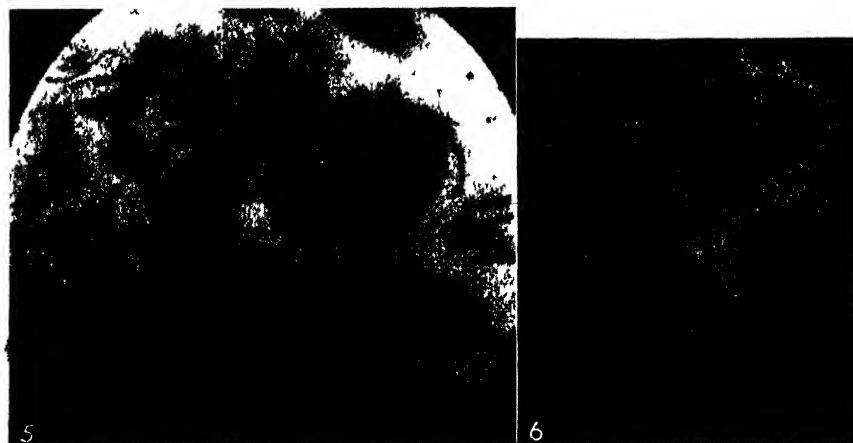
Fig. 5. Cross section of ampullae testis in triploid. Sperms and pycnotic nuclei. Gentian violet. Photomicrograph. (× 700). — Fig. 6. Cross section of ampullae testis in diploid. Gentian violet. Photomicrograph. (× 70).

triploid. It will be noticed that there are seven trivalents, four bi-valents and seven univalents, these being equivalent to thirty-six chro-mosomes (the number of chromosomes in a normal diploid is 24). For comparison Fig. 2 shows the corresponding stage in the diploid with twelve bivalents.

It is quite natural that the reduction division in this triploid should give rise to pre-spermatids with highly variable chromosome numbers (cf. Figs. 3 and 4). Just as a reduced pollen fertility is found in tri-ploid plants, a similar sterility might be expected here in the shape of a partially reduced sperm-cell viability. In spite of the fact that so far no sure cases of selective gametic elimination are known in animals, it seems highly probable that in the present material a number of pre-spermatids and spermatids would degenerate and be resorbed, owing to a highly deviating chromosomal constitution (cf. Fig 5). A com-parison with the diploid shows that the number of ripe sperms in the ampullae testis is substantially smaller (Figs 6 and 7)

So far as the sections show, the ampullae testis also contain typical degeneration products from spermatids that, probably on ac-count of changes in the genome, have not been able to complete their development into sperm-


Fig 7   Cross section of ampullae testis in triploid The number of histologically normal sperms in the ampullae is lower in comparison with Fig 6   Several pyc-notic nuclei can be observed   Gentian violet Photomicrograph — (× 70)

atozoa. There may accordingly have been present a haplontic sterility (MÜNTZING, 1930).

The size of the cell nuclei at the beginning of the first prophase as measured by their diameters shows an increase in the triploid by about 30 per cent. As yet, however, only a comparatively small num-ber were measured.

Those nuclei which are formed after the first division, correspond-ing to the pre-spermatids in the diploid, exhibit an immense variation in size. (It may be an open question if a nucleus, for instance formed by two or three univalents should be called a pre-spermatid or not.) This variation depends of course on the varying number of chromo-somes (cf. Figs. 8, 9 and 10). In certain cases it was also found that

three nuclei are formed at the first telophase. In some cases they are
of the same size, in others a few univalents form micronuclei. The
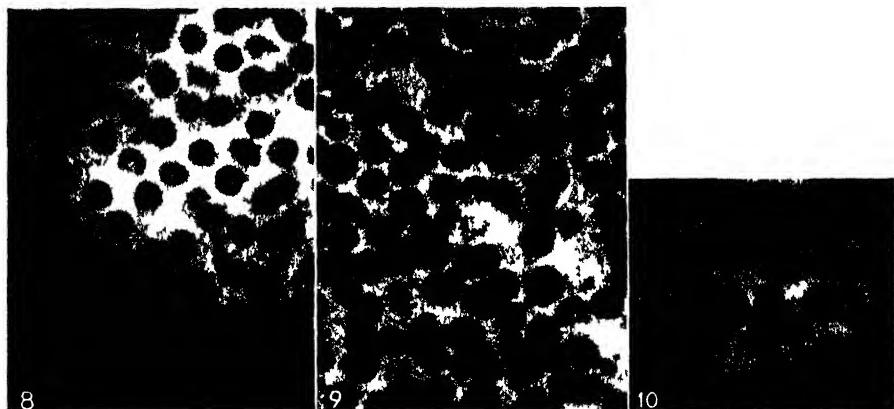
Fig. 8. Reduced nuclei of diploid testis. Very small variation in size Gentian violet
Photomicrograph. ($\times$ 350). — Fig 9. Reduced nuclei of the triploid testis. The
nuclei are on the average larger than in Fig 8 and there is a considerable variation
in size. Gentian violet. Photomicrograph ($\times$ 350) — Fig 10 Some reduced nuclei
of the triploid testis, showing very clearly the variation in size Gentian violet
Photomicrograph. ($\times$ 350).

Fig. 11. Metaphase plate of mitosis in the triploid Epithelial cell of epididymis
36 chromosomes ($\times$ 2000). — Fig. 12. Photomicrograph of the same plate drawn
in Fig. 11. Gentian violet. ($\times$ 550).

nuclei of the ripe sperms, i. e. the heads, also exhibit a corresponding
variation.

A complete mitotic metaphase plate, which is necessary for an

exact count of the chromosome number, was found in an epithelial cell of the epididymis (Figs. 11 and 12). In this exactly thirty-six chromosomes could be counted.

Sex. — Both the testes as well as the other sexual organs were macroscopically of the same size and appearance as in normal diploid males. What this implies from the point of view of sex-determination cannot be stated at present, since the sex-determining mechanism in amphibians is not yet known, although, as FANKHAUSER says, »there are indications that the male sex is heterogametic». Anyhow, the mechanism does not seem to be the same as in *Drosophila* (BRIDGES, 1922). If it were, triploids of the XXX and XXY types would be expected, the former being females (3X : 3A) and the latter intersexes (2X : 3A). No normal males could develop. Now in this species there is a male, undoubtedly uniformly triploid, without signs of intersexuality. Moreover, FANKHAUSER (1938 c) in the species *Triturus viridescens* described one individual which prooved to be triploid throughout and contained typical testes.

Fig. 13. First metaphase groups in triploid testis. Trivalents and univalents can be observed. Gentian violet. Photomicrograph. — ($\times$ 370).

The facts so far known, however, cannot explain the mechanism of sex-determination in this genus. It is possible that investigations on the sex-difference in triploid individuals, experimentally produced by cold treatment of fertilized eggs (see below and FANKHAUSER, 1939 b) may throw light on this problem.

Another line of approach is available in the investigation of the sex-difference in haploids. Of these, however, as yet only one animal has been developed so far that the sex could be established, and this was a female (FANKHAUSER, 1938 b).

The possibility of the sex-determining mechanism being phenotypically controlled must be pointed out, although this may be presumed to be a less likely contingency.

General discussion. — The occurrence of a probably partially fertile triploid vertebrate in a natural population must be awarded some significance from the points of view of species formation and morphogenesis. The assumption of the partial fertility of this individual is so far only based on the histological conditions. Since spermatohistogenesis inherently possesses a very balanced physiology, there would appear to be scarcely any reason to assume that histologically fully normal sperms should not be capable of function. In cases of complete sterility, e. g. in hybrids, no sperms at all are developed. The spermatocyte stage is not passed (e. g. *Carina* $\times$ *Anas*). On the other hand, when mature sperms are discovered in the testes of hybrids, these hybrids are also found to be partially fertile (e. g. *Serinus serinus* $\times$ *S. canarius*; POLL and TIEFENSEE, 1907).

Phylogenetically it is in principle immaterial whether the individual under investigation was a triploid throughout or not. It is extremely probable, though, that such was the case, especially in the light of FANKHAUSER's (1939 b) demonstration that triploid animals can be experimentally produced by means of cold treatment of normally fertilized eggs at the beginning of their development. It is to be assumed that this treatment prevents the formation of the second polar body, which normally is not formed until after the sperm has entered the egg, and the result is a triploid zygote.

This mode of formation may be assumed with a high degree of probability to be responsible for the origin of triploid animals in natural populations of the *Triton* species in this country. They lay their eggs early in spring, often while there is still ice in the water. Hence a severe night frost is all that is needed to create the same conditions as prevail during the experimental cold treatment.

Of course, it is also conceivable that a diploid egg-cell may arise in some other way than by external action, and at normal fertilization give rise to triploidy. Thus, for instance, chromosome bridges were twice observed in normal diploids, and one univalent was in one case observed, all during the first telophase of spermatogenesis. These three findings, however, are not sufficient as a basis for any theoretical explanation. Further investigations must, if possible, show the occurrence of meiotic disturbances that could give rise to diploid gametes.

Another conceivable explanation is the formation of unreduced sperm-cells, but these would then carry a relatively large amount of chromatin, and would probably not be able to compete successfully with the normal haploid sperms.

Lastly, with reference to polyspermy (dispermy), a physiological fertilization of this kind is of common occurrence in the *Triton* genus (cf. FANKHAUSER, 1924), and a pathological one with accompanying multipolar configurations at divisions could, presumably, scarcely lead to the origin of a triploid individual.

Of these different theoretical possibilities, the first (lowered temperature) appears to me to be the most probable. If this assumption is correct, a greater frequency of triploids ought to be found in regions where the climate affords the above-mentioned conditions. Further investigations will be conducted with a view to establish the frequency of triploids in nature.

As regards the *Urodela*, it seems as if triploidy has no considerable influence on their viability. In any case it does not constitute any obstacle to the attainment of sexual maturity. A further increase of the chromosome number to tetraploidy, however, results according to FANKHAUSER (1939 b) in a reduction of size and a considerable lowering of viability. This would suggest that the animal organism is much more sensitive than the plant organism to polyploidy.

According to MÜNTZING (1936), the occurrence of polyploid series among plants is very common. However, no direct homology to this has been discovered in animals, which might be explained on the above-mentioned grounds. The abundant occurrence in triploids of gametes with varying chromosome numbers indicates, however, a path by which the chromosome number could undergo an increase. The previous reference to haplontic sterility did not imply that all gametes with aberrant chromosome numbers are eliminated by selection, but probably only the most extreme ones. Crosses between diploids and triploids may reveal whether this possibility has any foundation in fact. In plants, however, individuals with extra chromosomes are known to have a reduced viability (cf. DARLINGTON, 1937).

Another fact of phylogenetic importance that stands out rather clearly is that triploids by virtue of the meiotic disturbances which must occur in them offer greater chances for the origin of new chromosome types. In plants, for instance, it has been shown that the frequency of structural chromosome changes is considerably increased in triploids (cf. MÜNTZING, 1939, p. 343).

Appendix. — It was also found that the nuclei of the erythrocytes were definitely larger. This indicates that at least the whole mesoderm must have been triploid. On the basis of this fact, also found by FANKHAUSER in his material, a method to identify an increased chro-

mosome number by blood diagnosis is under elaboration. If this
method really works it might be extended even to other vertebrate
classes.

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FURTHER STUDIES ON APOMIXIS AND SEXUALITY IN POA

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INTRODUCTION.

SEVEN years ago the present writer reported the occurrence of apomictic seed development in *Poa pratensis* and *Poa alpina* (MÜNTZING, 1932), and in the same paper the occurrence of a sexual *alpina* strain was also described. The chromosomal conditions in the material studied were found to be peculiar, the biotypes of both species being characterized by various, mostly aneuploid, chromosome numbers.

These findings have been verified and much extended by several workers (NILSSON, 1933, 1937; RANCKEN, 1934; KIELLANDER, 1935, 1937; AKERBERG, 1936 a, 1936 b, 1938, 1939; FLOVIK, 1938).

Though progress in my own *Poa* investigations has been slow, several data have accumulated, which justify the following report. The results chiefly concern the occurrence and inheritance of apomixis in *Poa alpina*, but some observations gathered in *Poa pratensis* may be recorded at the same time.

I. THE APOMICTIC BIOTYPES IN POA ALPINA.

1. MATERIAL.

Originally, eight apomictic *alpina* biotypes were studied. Six of these are of Swedish origin, the other two were obtained from Norway and Switzerland. Two of the Swedish biotypes are from Korpilombolo and Pajala in North Sweden and have the chromosome numbers 38 and 33 respectively. A third biotype from Jämtland (Middle Sweden) was found to be characterized by the chromosome number 38. Of two south Swedish biotypes the one from Mösseberg has 2n = 33, whereas the other one, from the island of Öland, was found to have 2n = 35. Of the foreign apomictic *alpina* biotypes the one from Norway (Troms) was found to have 38 chromosomes, like the types from Korpilombolo

and Jämtland. Finally, the Swiss biotype (from St. Gothard) represented a new number, viz. 2n = 37.

In addition to these 8 strains, different degrees of apomictic seed development were also observed in a new collection of Swiss *alpina* types. This material, however, will be discussed below under a separate heading.

2. CHROMOSOMAL AND MORPHOLOGICAL ABERRATIONS.

Chromosome counts have revealed the fact that some of the strains studied are not absolutely constant, a certain proportion of individuals with deviating chromosome numbers being produced. This is evident from Table 1, which summarizes all chromosome numbers so far obtained in the material in question. In strains 3, 4 and 7, the values given in the table were all found in a single individual progeny after isolation, in the other strains the values represent the total result from counts of more than one progeny (cf. below p. 118). In each strain, however, the different progenies gave quite similar results.

The highest degree of aberrant formation is found in strain No. 8 (St. Gothard). In this strain (Fig. 17) the chromosome numbers of 27 individuals were determined, and of these plants 16 were found to have 2n = 37. Evidently, this is the typical number of the strain, the remaining 11 plants having more or less clearly aberrant numbers. Of these aberrants those with 45, 67, 72 and 74 chromosomes are quite indisputable, but the other deviations from 37 may be suspected to be due to slight errors in counting. The plant with 2n = 33, however, is certainly a true aberrant, three rather clear chromosome plates giving the same number. The plants having presumably 36, 38 and 39 chromosomes are more dubious, and the evidence that they really differ from the typical number 37 is not quite convincing.

Morphologically, the aberrants with 45 to 74 chromosomes had been recognized to be more or less clearly deviating before the chromosome numbers were known. The other plants, including the one with 2n = 33, were not seen to show any morphological deviations. — The six aberrants with high numbers were less vigorous than the typical plants. Vigour in this material was estimated by using a scale from 1—10, the higher values representing the more vigorous plants. The values of the six aberrants in question ranged from 3 to 5, the average being 4,5. The corresponding values of ten typical plants, having 2n = 37, ranged from 6 to 9 with an average of 8,2.

The typical chromosome number of the Gotland biotype (Fig. 15)

is evidently $2n = 33$, this number being found in 11 out of 15 plants studied (Table 1, strain 7). The other chromosome numbers are clearly deviating ($2n = 41, 49, 64$ and 65 respectively). An interesting point is that these aberrants were not detected by morphological inspection. Also the vigour of the aberrants was in this case equal to that of the plants with a normal chromosome number.

Two other strains have also produced aberrants with high chromosome numbers, viz. the Troms and Mösseberg strains. In the former a plant with 74 chromosomes appeared, whereas the typical number of the strain is evidently 38. In the Mösseberg strain 44 out of 46 plants were found to have $2n = 33$. Of the two aberrants one had $2n = 52$, the other $2n = 28$. The latter plant died before morphological notes were made, the other aberrant with $2n = 52$ had quite good vigour but was not observed to be morphologically divergent. The aberrant plant in the Troms strain, on the contrary, was weaker than the normal plants and was conspicuous by its markedly bluish green leaves, stiff panicles and short panicle branches.

In the remaining four strains (Nos. 1, 2, 3 and 8) no true aberrations seem to occur. The values slightly differing from the typical numbers (38, 33, 38 and 35 respectively) are probably due to slight errors in counting. Thus, for instance, in one of the plants belonging to strain 3 two of the three plates studied gave the value 37, while the

TABLE 1. *Chromosome numbers in some apomictic strains of Poa alpina.*

Strain number	Origin	Somatic chromosome numbers																				n
		28	33	34	35	36	37	38	39	40	41	45	49	52	64	65	66	67	72	73	74	
1	Korpilombolo, Sweden						1	20	7													28
2	Pajala, Sweden		26	5																		31
3	Jämtland, Sweden						1	6														7
4	Troms, Norway						1	5													1	7
5	Mösseberg, Sweden	1	44											1								46
6	Öland, Sweden			1		32	5									1						39
7	Gotland, Sweden		11								1		1		1	1						15
8	St. Gothard, Switzerland			1	16	1												1		3	1	27

third plate seemed to contain 38 chromosomes. Therefore, although the chromosome number of the plant was considered to be $\pm$ 37 and given as 37 in the table, it is possible and even probable that the correct number is 38, the typical number of the strain under consideration. The same arguments apply to all other deviations by $\pm$ 1 from the typical number. Even an apparent deviation by 2 chromosomes may possibly be due to difficulties in counting, especially if the fixations are not very good. Thus, in strain 6 there is one value of 37 in the table. This count was based on four plates, giving the values 37, 37, 37 and 35. Since 35 is the normal number in this strain it is not excluded that 35 is the correct number.

Summing up, it can be said that *four of the eight strains studied seem to be perfectly constant in chromosome number, and this is also accompanied by a complete morphological uniformity. In the four other strains a certain proportion of true aberrants with a slightly deviating morphology are produced, but the frequency of aberrants differs in different strains.* Thus, in strain 8 there are at least 7 aberrants, which corresponds to a percentage of $25{,}93 \pm 8{,}43$. In strain 5 the corresponding percentage value is $4{,}35 \pm 3{,}01$. The difference is $21{,}58 \pm 8{,}95$ and $D/m = 2{,}41$. The odds that this difference is significant are 63 : 1. It should be remembered that the percentage of aberrants in strain 8 is a minimum value, the true value probably being somewhat higher. Under such circumstances the significance of the difference in question is beyond reasonable doubt. — If a higher number of individuals had been studied it is quite possible that a few aberrants would have appeared also in the strains that now seem to be perfectly constant. At any rate the percentage of aberrants must, however, be very low in these strains.

It is striking that most of the aberrants formed have an increased number of chromosomes. In the table there is a total of 14 indisputable aberrants, and of these only two have lower numbers than normally. Of the 12 aberrants with high numbers 8 plants are exactly or approximately tetraploid in relation to the normal plants, two aberrants are approximate triploids, and the remaining two represent other deviations.

As already mentioned above, the chromosome numbers of biotypes 1, 2, 5, 6 and 8 cited in Table 1 represent the sum of chromosome counts in more than one progeny. These progenies were partly raised after isolation, partly after open pollination. Since it was found, however, that in progenies of both kinds the chromosome numbers were

quite similar only the total values are given in Table 1. It should be mentioned, however, that exactly half of the plants studied were raised from a crossing group consisting of one plant each of the strains 1, 2, 5, 7 and 8. These plants were put together in an isolation cage in the greenhouse, and clouds of pollen were repeatedly induced inside the cage by rubbing the plants with a stick. This experiment was undertaken in order to see, whether a rich access to foreign pollen would diminish the constancy of the biotypes, previously observed. If the results of isolation and mixed pollination are compared the following survey is obtained:

	Isolation		Mixed pollination	
Strain No.	Typical plants	Aberrants	Typical plants	Aberrants
1	6	—	22	—
2	7	—	24	—
3	7	—	—	- -
4	6	1	— -	—
5	27	1	17	1
6	39	-	— -	—
7	—	-	11	4
8	6	-	14	7

Evidently the mixed pollination did not increase the number of aberrants in biotypes 1, 2 and 5. Only in biotype 8 do we find any indication of a positive result, all 7 aberrants obtained occurring in the progeny from mixed pollination.

3. THE TYPICAL CHROMOSOME NUMBERS.

Concerning the typical chromosome numbers of the strains studied, it is interesting to note that with the exception of the Öland strain, which has $2n = 35$, all the other Scandinavian strains have either 33 or 38 chromosomes. The number 38 is evidently typical of the strains from Korpilombolo (in the province of Norrbotten) and Jämtland as well as of the Norwegian strain from Troms. As reported previously (MÜNTZING, 1932), the same number was also found in a biotype from Pajala (in the same province as Korpilombolo). The Pajala biotype considered in the present paper, however, has $2n = 33$, and the same number is present in the Mösseberg biotype and also in the biotype from Gotland. These three localities are separated by wide distances. Nevertheless, *this identity in rather peculiar chromosome numbers*

suggests some kind of relationship within the 38 chromosome as well as the 33 chromosome group. Morphologically, however, all the biotypes, even those having the same number, were clearly distinct.

The biotype from Öland is rather different from the other Scandinavian *alpina* biotypes, not only in its chromosome number (2n = 35) but also by the fact that it is quite difficult to keep in culture. Several progenies of this biotype have been raised, but almost all plants in these progenies only survived one summer and died during the following winter. Thus, in contrast to the other Scandinavian *alpina* types, the Öland apomict studied is ephemerical. It remains to be studied whether this type also behaves as an annual in its proper habitat.

4. FERTILITY.

As already reported in my previous *Poa* paper, pollen fertility in the *Poa alpina* apomicts is quite good. All data on pollen fertility available are summarized in the following table:

Strain No. (cf. Table 1)	Per cent apparently good pollen							n	M
	40 —	50 —	60 --	70 --	80 —	90 —	100		
1							7	7	95,0
2				1	11	22	34		91,2
5	1	1	2	2	1	1		8	70,0
6			2	6	11	3		22	81,8
8				1	3	4		8	88,8

Strains 5 and 6 (Mösseberg and Öland) seem to have less good pollen fertility than the other strains, but more data are needed in order to prove that definitely. The main thing, however, is the high average pollen fertility, which is just as good as in any sexual cross-breeding plant species (cf. MÜNTZING, 1939).

In all strains observed seed production was found to be abundant upon open pollination, and no difficulties were encountered in getting plenty of seeds on isolated panicles. — Only in one single case was a failure of seed setting met with. This was in a pot plant of the Gotland strain which flowered alone in a relatively dry and warm greenhouse. For some reason most of the anthers of this plant did not dehisce, and very few seeds were produced in spite of the fact that the female organs seemed to be quite normally developed, and the plant was typical morphologically. In another plant of the same strain, flowering at the

same time in an isolation cage in the same greenhouse, pollen as well as seed production were found to be normal. Thus, though no direct crossing experiments have been undertaken, the observation made
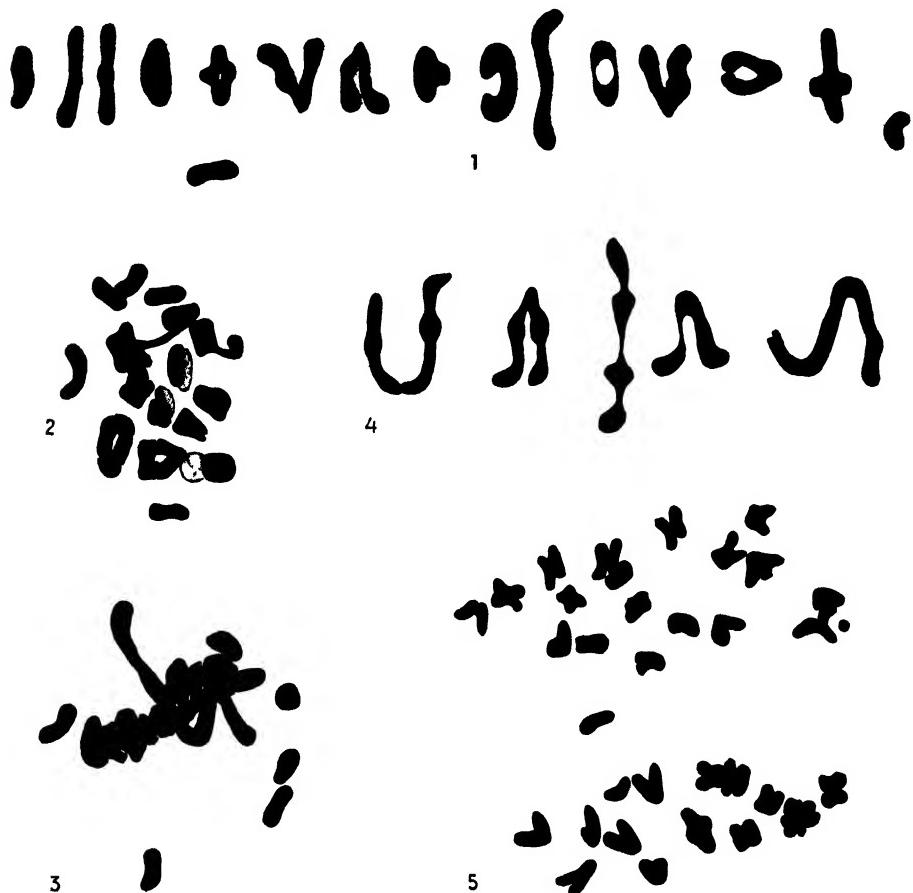
Figs. 1—5. Meiosis in the apomictic *Poa alpina* biotype from Pajala, North Sweden $(2n = 33)$. — Fig. 1, I—M in side view (separately drawn), $3_{III} + 11_{II} + 2_I$; Fig. 2, I—M in polar view, probably $2_{IV} + 2_{III} + 7_{II} + 5_I$; Fig. 3, I— M group with multivalent and 7 univalents; Fig. 4, multivalents from different I—M groups (the one to the right not visible in its entire length); Fig. 5, I—A with the distribution 18—2/2—14.

strongly indicates that pollination is necessary for seed development in the strain in question. Especially since ÅKERBERG (1936 a) has proved *Poa pratensis* to be pseudogamous, it is quite probable that apomixis in *Poa alpina* is of the same kind.
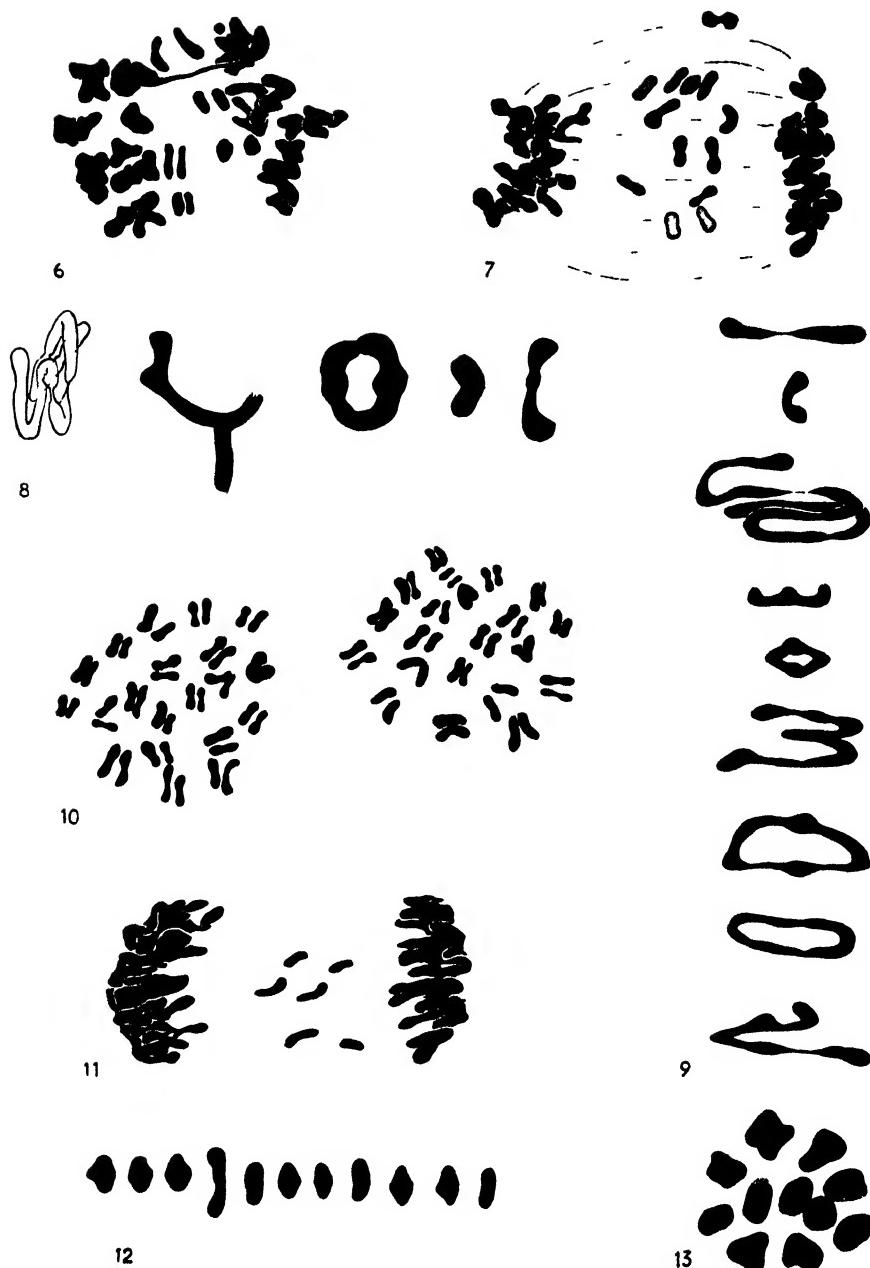
Figs. 6—7. Meiosis in the Pajala apomict of *Poa alpina* (continued). — Fig. 6, I—A with dicentric chromatid and fragment, 5 univalents divide; Fig. 7, I—A showing division of 7 univalents. — Fig. 8, I—M associations in the apomictic *alpina* biotype from Mösseberg (2n = 33); from the left to the right: a multivalent

5. MEIOSIS.

Meiosis in the p. m. c. was mainly studied in one of the apomictic *alpina* strains, viz. the Pajala strain with $2n = 33$. Some additional evidence was also gathered from the Mösseberg and Korpilombolo strains.

In the Pajala strain meiosis is rather irregular and may be characterized as follows: At I—M there are plenty of univalents which generally divide at I—A and lag at II—A. As a consequence of these irregularities there is a considerable amount of chromosome elimination. Of the I -M associations bivalents are most frequent, but trivalents also occur. Associations of more than three chromosomes are more rarely met with. At meiosis no fragments were observed, the univalents being of normal size. *The strain in question may evidently be characterized as a quite unbalanced, partially autopolyploid, aneuploid.* This characterization is based on the following detailed observations (cf. Figs. 1—8):

At I—M a total of ten complete groups were found to represent the following eight configurations:

$$2_{IV} + 2_{III} + 7_{II} + 5_{I} \text{ (1 cell)}; \quad 3_{III} + 11_{II} + 2_{I} \text{ (1 cell)}$$
$$1_{IV} + 2_{III} + 10_{II} + 3_{I} \text{ (» »)}; \quad 3_{III} + 10_{II} + 4_{I} \text{ (2 cells)}$$
$$1_{IV} + 1_{III} + 10_{II} + 6_{I} \text{ (» »)}; \quad 3_{III} + 9_{II} + 6_{I} \text{ (» »)}$$
$$1_{IV} + 12_{II} + 5_{I} \quad \text{(» »)}; \quad 1_{III} + 13_{II} + 4_{I} \text{ (1 cell)}$$

Thus, in all cells studied trivalents or quadrivalents or both were present. The maximum number per cell of such associations is 4 and the average number is $2_{,6}$. Since the fixation was not ideal, it is possible that the interpretation is not always accurate, but in the main the configurations given are certainly correct. Two of the configurations are represented in Figs. 1—2. In Fig. 1 there are 3 trivalents, 11 bivalents and 2 univalents, in Fig. 2 the configuration is probably $2_{IV} + 2_{III} + 7_{II} + 5_{I}$. Most of the trivalents are V-shaped. Some separate trivalents and quadrivalents are represented in Fig. 4. In Fig. 3 the

composed of 7 or 8 chromosomes, part of a multivalent, a quadrivalent and (for comparison) two bivalents from the same group. — Figs. 9—11, I—M and I—A in an F_1 hybrid of the cross sexual × apomictic *Poa alpina* $(2n = 41)$. — Fig. 9, five multivalents and for comparison three bivalents and one univalent. The bivalents and the univalent are from the same I—M group as the large multivalent, which is probably composed of seven chromosomes; Fig. 10, I—A plates, showing the distribution 20—21; Fig. 11, I—T, division of three univalents. — Figs. 12—13, regular I—M in a sexual *Poa alpina* plant with $2n = 22$. Fig. 12, side view (separately drawn), 11_{II}; Fig. 13, polar view, 11_{II}.

metaphase group is characterized by the presence of a big multivalent, consisting of at least four chromosomes. In the same group there are as many as 7 univalents
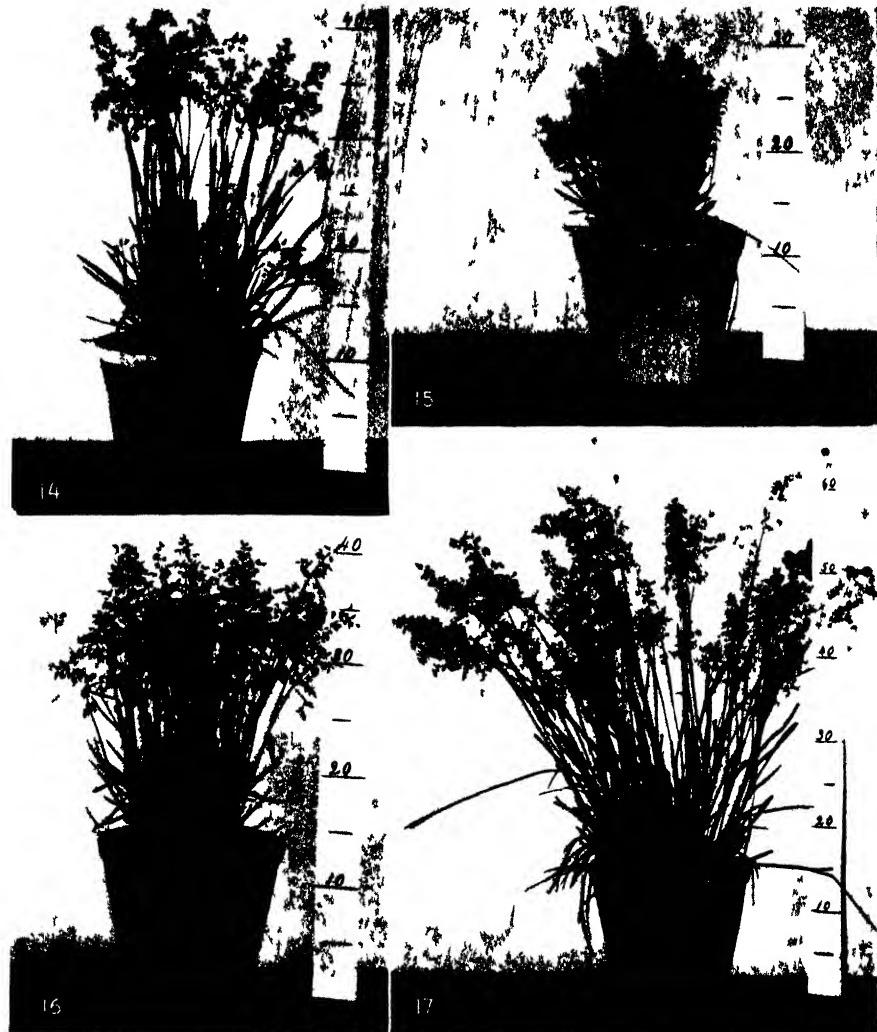
Figs 14—17 Four different *Poa alpina* apomicts — Fig 14 represents the biotype from Mosseberg (South Sweden), Fig 15 the biotype from the island of Gotland, Fig 16 the biotype from Pajala (North Sweden), and Fig 17 the strain from St Gothaid (Switzeiland)

The number of univalents at I—M was counted in 50 cells, the following distribution being obtained:

Number of univalents: 0 1 2 3 4 5 6 7
»   » cells:.....   3 7 13 14 7 5 1

Thus, univalents were present in every cell observed, their average number being 3,68.

At first anaphase dividing univalents were observed in every cell (cf. Figs. 6—7). The frequency was found to be as follows:

Number of dividing univalents: 1   2   3 4 5 6 7
»   » cells: 2 10 13 8 5 4 1

The average number is 3,47, which closely corresponds to the average number of univalents present at I—M.

Fig. 5 shows a first anaphase (somewhat flattened by pressure) in which the distribution is 14—2/2—18. In addition to these chromosomes there is a possible fragment in the upper group.

A quite clear case of dicentric chromatid and fragment is visible in Fig. 6. Thus, the strain in question must be heterozygous for an inversion or a duplication.

In addition to the data gathered in the Pajala biotype, some observations could also be made in the Mösseberg strain, which has the same chromosome number (2n = 33). At I—M the chromosome configurations are evidently of the same type as in the Pajala strain. Fig. 8, third association from the right, represents a clear quadrivalent. For comparison two bivalents from the same group are drawn to the right of this quadrivalent. In Fig. 8, to the left, there are two other multiple configurations. One of them (drawn in outline) consists of at least 6 chromosomes. The other big multivalent is also composed of several chromosomes but is only partly visible.

Meiotic irregularities occur not only in the apomicts with 2n = 33 but were also observed in one of the strains with 38 chromosomes (the Korpilombolo apomict). Also this strain was found to be characterized by the occurrence of univalents and multivalents, besides bivalents, at I—M and showed the usual resultant irregularities at later stages. The number of univalents at I—M was counted with the following result:

Number of univalents: 0   1   2   3 4 5
»   » cells:..... 4 10 24 12 1 2

The average number is 2,04 and seems to be lower than the corresponding value, 3,68, obtained in a slide of the Pajala apomict (2n = 33).

6. EMBRYOLOGY.

The occurrence of apomixis in *Poa* was first demonstrated by cytogenetic methods, but, naturally, an embryological verification of the results obtained seemed highly desirable.   Therefore embryological fixations of some *alpina* and *pratensis* strains were made.   During a stay in Stockholm in 1933 I had the opportunity of making a preliminary study of this embryological material under the guidance of Pro-
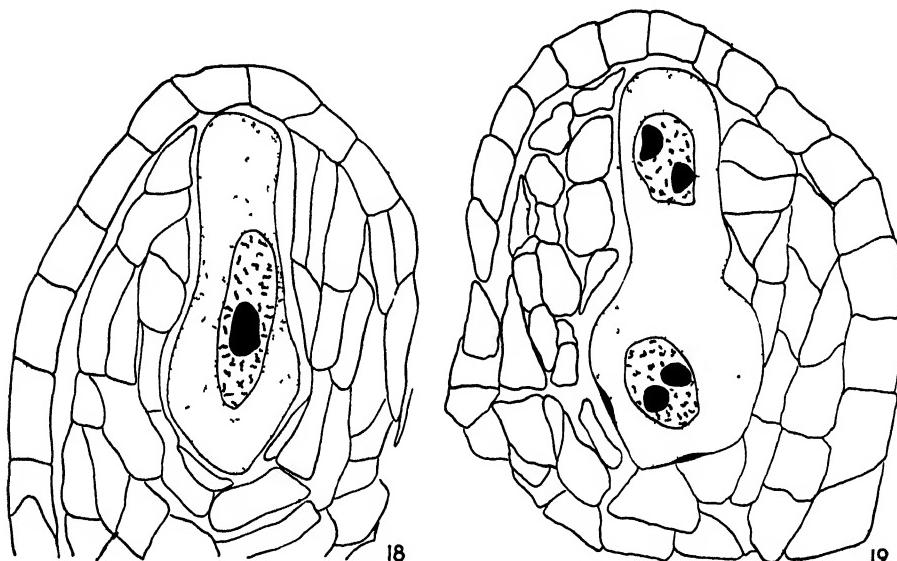
Figs 18—19   The first stages of embryo-sac development in an apomictic *Poa alpina* biotype — Fig. 18, the macrospore mother cell is enlarged and contains several vacuoles, Fig 19, a developing embryo-sac with two nuclei, no degenerating macrospores

fessor O. ROSENBERG.   Though, for various reasons, this investigation was never finished one result obtained may be mentioned.

In the Pajala strain of *Poa alpina* (2n = 33) the first stages of embryo-sac development were observed (Figs. 18—19).   Without exception the embryo-sac was found to develop directly from the macrospore mother cell, meiosis thus being omitted.   Fig. 18 shows a uninucleate macrospore mother cell, which is evidently passing through the first stages of embryo-sac development.   This is evident from the conspicuous vacuolisation.   In Fig. 19, the next stage, a binucleate embryo-sac is met with.   At this stage no trace of degenerating macro-

spores can be seen, thus confirming the conclusion that we have here a case of *diploid parthenogenesis*, according to the *Antennaria*-scheme.

II. THE SEXUAL POA ALPINA MATERIAL FROM SWITZERLAND.

1 THE FIRST PROGENIES.

A CHROMOSOMAL VARIATION

As described in the previous paper (MÜNTZING, 1932), a seed sample of *Poa alpina* obtained from Switzerland gave rise to a material
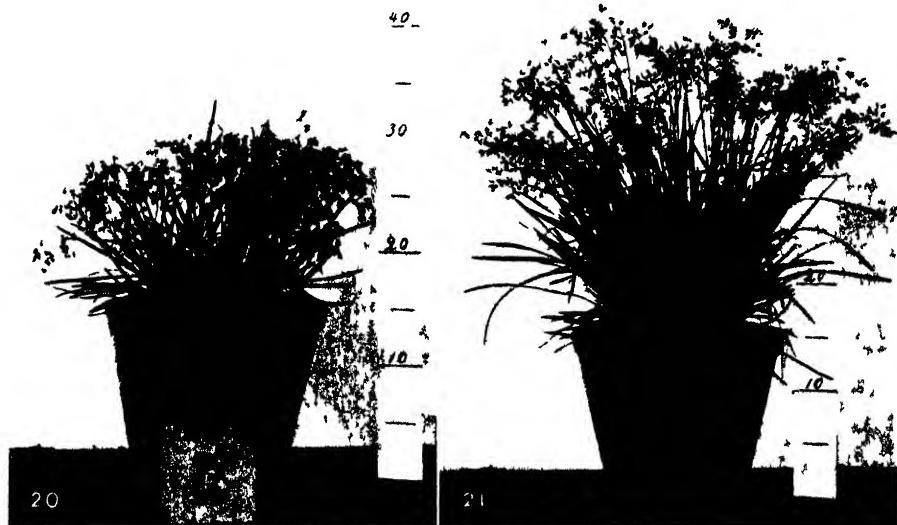
Figs 20 21   Two plants of the sexual *Poa alpina* strain from Furstenalp, Switzerland The plant in Fig 20 has 31 chromosomes, the individual in Fig 21 has 2n — 22

which was highly variable in chromosome number as well as morphology (cf. Figs. 20—21). In order to prove that this variation was due to sexuality, three progenies were raised from isolated mother plants, having the chromosome numbers 24, 25 and 31 respectively. Chromosome counts in a total of 27 daughter plants revealed the variation given in Table 1 of the paper mentioned. Since that was written a higher number of plants in these progenies have been examined cytologically and chromosome numbers are now available for 77 individuals. The chromosomal variation among these plants is given in Table 2.

TABLE 2. *Chromosome numbers in progenies of sexual Poa alpina plants.*

Field number	Somatic chromosome numbers													n	M	Chromosome number of the mother plant
	21	22	23	24	25	26	27	28	29	30	31	32	33			
32—52	1	4	5	3	2	1								16	23,25	25
—53		9	9	14	1	5	—	1						39	23,69	24
—55							1	4	3	5	3	2	4	22	30,23	31

The following conclusions may be drawn from the table: a) all three mother plants must be sexual; b) there is a slight average decrease in chromosome number, and the aberrants with lower numbers than the mother are more numerous than those with higher numbers.

Thus, the mother plant having $2n = 25$ gave a progeny with the average chromosome number 23,25, the corresponding values of the other two mother plants being 24—23,69 and 31—30,23. This average decrease is probably due to meiotic irregularities and chromosome elimination. It may be observed, however, that the degree of this elimination may differ in different plants. Thus, the mother plant 32—53 is evidently more stable than the 32—52, judging from a lower degree of chromosome variation in the progeny and a very slight decrease of the average chromosome number. Even in this progeny, however, the number of aberrants with lower chromosome numbers than the mother is clearly higher than the aberrants with higher numbers (18 versus 7).

B. CORRELATION BETWEEN CHROMOSOME NUMBER, VIGOUR AND FERTILITY.

In the three progenies just discussed the chromosome numbers vary from 21 to 33, all intermediate numbers also being represented. Since variation in viability and fertility was obvious in this material, it seemed desirable to test whether correlation could be found between chromosome number, vigour and fertility. Starting with chromosome number and vigour (Table 3) it is surprising to find that these variables do not show any obvious correlation.

Vigour was estimated with the aid of a scale from 1 to 5, the latter figure corresponding to the most vigorous plants. In the vigour classes 1—5 the average chromosome numbers were found to be 22,6;

TABLE 3. *Correlation between chromosome number and vigour in a sexual strain of Poa alpina.*

Vigour	Somatic chromosome numbers													n	M
	21	22	23	24	25	26	27	28	29	30	31	32	33		
1	1	1	2	1										5	22,6
2		1	1	3	2	1	—	1	—	1	—	—	1	11	25,8
3		3	2	4	1	2	1	—	—	3	—	3		19	26,2
4		5	6	6	1	—	—	2	1	1	1			23	24,5
5		1	2	3	-	2	—	1	—	—	—	1		10	25,2

25,8; 26,2; 24,5 and 25,2 respectively. The absence of a correlation might be suspected to be due to the fact that the material is heterogenous, consisting of three different progenies. However, attempts to find a correlation within each progeny were equally unsuccessful. Thus, the only conclusion to be drawn is that weak and vigorous plants are present in all chromosome classes in about the same frequency.

Similar attempts to find a correlation between chromosome number and pollen fertility gave about the same result (Table 4). In all chromosome classes there is a high proportion of quite male sterile individuals with non-dehiscing anthers, and among the pollen producing plants several were found to be partially sterile. The plants having the lowest chromosome numbers (21—23) seem to have less good pollen than the other plants, but the number of individuals is too low to furnish definite proof.

When studying fertility and vigour in this material it was observed that the male sterile plants with non-dehiscing anthers were characterized by their poor tillering, whereas plants with many panicles

TABLE 4. *Correlation between chromosome number and fertility in a sexual strain of Poa alpina.*

Chromosome numbers	Per cent good pollen						n	M	Quite sterile	Per cent quite sterile
	50	60	70	80	90	100				
21—23	1	5	3	2	2		13	74,2	12	48
24—26				2	9		11	93,2	15	58
27—29				2	3		5	91,0	1	17
30—33			1	2	2		5	87,0	7	58
Total	1	5	4	8	16		34	84,7	35	51

generally had better fertility. Thus, a quite clear positive correlation was found between degree of tillering and male fertility, plants with few panicles often having defective anthers. This is evident from the following values:

	Degree of tillering				
	1	2	3	4	5
Fertile:	5	4	7	4	14
Sterile:	12	5	—	2	2

More than half the »sterile» plants with non-dehiscing anthers belong to the poorest tillering class (1), while the maximum class of the »fertile» plants (producing pollen) corresponds to the best tillering class (5). The significance of the difference in distribution is obvious without statistical treatment.

2. SELECTION FOR HIGH AND LOW CHROMOSOME NUMBERS.

According to the data given above, *the chromosome number in the sexual material under discussion is oscillatory and without any clear effect on fertility and viability.* Under such circumstances it seemed desirable to increase and decrease the chromosome number by selection as far as possible. The selection for high numbers was also undertaken with the possibility in mind that apomictic strains might be produced in this way, the apomictic strains occurring in nature being characterized by a higher average chromosome number than the present sexual material.

A. SELECTION FOR HIGH CHROMOSOME NUMBERS.

In the sexual progenies represented in Table 2 most of the plants have the same or lower chromosome numbers than the mother individuals. This is also true of the progeny 32—55. However, in this, as in the other progenies, a few plants had higher numbers than the mother. These plants, having $2n = 32$ or 33, and some other plants with at least 31 chromosomes, were isolated. Five of the isolated plants produced seed in the isolation bags and gave rise to the progenies represented in Table 5. One progeny (1935—9) consists of a single plant with $2n = 35$, raised from a mother with $2n = 33$. Two other progenies, 1933—12 and 1935—12, are also rather small but are sufficient to show that variation in the offspring goes in the plus as well as the minus direction. Most of the material, however, belongs to the progenies 1935—10 and —11, in which chromosome counts are available from a total of 102 plants. Due to an error in the work of pricking,

the progenies 1935—10 and —11 were partially mixed. Fortunately, the mother plants had the same chromosome number, 31, and thus the chromosomal variation in their total offspring is still of interest.

From the table it is evident that chromosomal variation in these progenies, from mother plants with high numbers, is just as great as in the original sexual material. Thus, so far at least, the selection for high numbers has by no means resulted in an apomictic tendency. It remains to be seen how the individuals with 34 to 64 chromosomes will behave in the next generation, but it does not seem very likely that their stability will be increased.

Though several plants with high chromosome numbers have appeared in the progenies under discussion, it is obvious that there is a much stronger tendency to decrease the chromosome number. Thus, in 1935—10 + 11 20 plants out of 102 had the same chromosome number as the mother plants (2n = 31), 57 plants had lower numbers and 25 plants had higher. If the values of the other progenies are added also, and the values are expressed in per cent, the result will be the following: *Of a total of 114 plants 19 per cent had the same number as the mother, 55 per cent had lower numbers and 26 per cent higher numbers.* At least part of the minus tendency is certainly due to meiotic irregularities and chromosome elimination.

Though the proportion of plus

TABLE 5. Chromosomal variation in progenies of sexual alpina plants with high chromosome numbers.

Field number	Chromosome number of the mother plants	Somatic chromosome numbers																							
		25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46 ... 57	... 64
1933—12	31					1	4	1	—	1				1	1					1			—	—	1
1935—12	32					1			1	2				1		1	1	1		1			—	1	—
1935—9	33						1				1	1										1	1	—	—
1935—10	31	2	2	2	3	5	7	11	4	4	1				1	1	1	1	1	1	1			—	1
1935—11	31		4	8	3	10	11	9	—	4		1		2		1		1		1		1		—	1
1935—10 + 11	31	2	6	10	6	15	18	20	4	8	1	1		2	1	1	1	1	1	1	1	1		—	1

variates is relatively low their degree of variation is much higher than
that of the minus variates. In the families 1935—10 and —11 the
numerous minus variates comprise only 6 chromosome classes (down
to 25), whereas the plus variates represent 14 different chromosome
numbers (up to 64). Undoubtedly this very wide plus variation is due
to the functioning of unreduced or approximately unreduced gametes.
If such ± unreduced gametes unite with reduced ones, which certainly
vary in chromosome number very much, a rather wide range of chro-
mosomal variation may be expected. Thus, the ten plants with 37 to
46 chromosomes probably originated in this way. — In some cases two
± unreduced gametes may be expected to unite, and this procedure
may be responsible for the two plants with 57 and 64 chromosomes.

In the progenies 1935—10 + 11 the chromosome numbers varied
from 25 to 64 and at the same time a conspicuous variation in vigour
was observed. However, also in this material no correlation between
chromosome number and vigour could be established. The material
was divided into the chromosome classes 25—27—29 . . ., vigour was
estimated by means of a scale from 0 to 10. The first value represents
plants found to be dead in the field, the latter value the most vigorous
plants. In the present material, however, the highest vigour value
reached was 7. The average vigour values in the different chromosome
classes were found to be the following:

Class	Average vigour	Number of individuals
25—27	4,0	8
27—29	3,3	16
29—31	3,7	33
31—33	4,3	24
33—35	3,9	8
35—64	3,0	10

There are no significant differences in vigour between the different
chromosome classes.

At the same time as the material just discussed, progenies of
two other categories were grown under the same conditions. These
categories were firstly five progenies of apomictic mother plants (the
Korpilombolo, Pajala, Mösseberg, Gotland and St. Gothard strains)
and, secondly, a material of sexual plants selected for low chromosome
numbers. As will be described below, most of these plants had 22
chromosomes.

It may be of some interest to compare the vigour of these three groups of plants. The following values were obtained.

Category	Vigour 0 — 2 — 4 — 6 — 8 — 10					n	M ± m
Apomictic strains:	6	32	153	22	16	229	5,08 ± 0,10
Sexual, high numbers: ...	24	22	69	7		122	3,96 ± 0,16
» , low » : ...	29	50	73	11		163	3,80 ± 0,13

On an average the sexual plants are evidently less vigorous than the apomictic strains. This is quite natural, since the apomicts are constant and successful selection products, the sexual strains, on the contrary, being highly variable in vigour. Thus, the occurrence of a considerable proportion of inferior individuals in the sexual strains is responsible for their relatively low average vigour.

In the sexual material the two groups with high and low chromosome numbers have about the same average vigour. This accords well with the results described above, which were obtained within the material selected for high chromosome numbers and the observations made in the primary sexual progenies (p. 128). It demonstrates once more a surprising degree of independence between vigour and chromosome number in this material.

B. SELECTION FOR LOW CHROMOSOME NUMBERS.

Already among the 9 plants raised from the original seed sample, giving rise to the sexual material, there were two individuals having $2n = 22$ (MÜNTZING, 1932, p. 133). Several new plants with 22 chromosomes appeared in the progenies 32—52 and 32—53 (Table 2) and even a plant with $2n = 21$ was obtained. The latter was poor in vigour as well as fertility, and no seeds from isolation could be obtained. Among the plants with $2n = 22$, however, isolation was more successful, and progenies were raised from eight different mother plants. Chromosome counts were undertaken in these progenies and the result is given in Table 6. Of the mother plants cited in this table Nos. 49, 50, 52 and 57 are sister plants, all of them belonging to progeny 32—52 (Table 2). Plants Nos. 75 and 82 are also sister plants and were obtained from progeny 32—53 (Table 2). Finally, the mother plants 143 and 144 in Table 6 represent one generation later and are daughters of plants Nos. 52 and 57 respectively.

The main result of the chromosome counts is rather striking.

Evidently, *the great majority of the plants have 22 chromosomes just as their mothers.* Considering the total values first, the chromosome numbers of 122 plants were determined, and of these not less than 103 (84 per cent) had 2n = 22, one single plant had 21 chromosomes and 18 plants were characterized by numbers higher than 22. Two of the latter values, viz. 31 and 33, probably resulted from the union of un-reduced and reduced gametes.

The progenies seem to represent different degrees of constancy. Taking the extremes, $22,2 \pm 6,2$ per cent of the plants in the offspring of plant 49 have chromosome numbers other than 22. The corresponding value in the progeny of plant 57 is $7,4 \pm 5,0$. Since the difference

TABLE 6. *Chromosomal variation in progenies of sexual alpina plants, having 2n = 22.*

Mother plant No.	Somatic chromosome numbers													n
	21	22	23	24	25	26	27	28	29	30	31	32	33	
49		35	5	2	1	1	—	—	—		1			45
50		1												1
52		31	4	1										36
143		6												6
57	1	25	—	1										27
144		2												2
75		3												3
82		2	—	—	—	1	—	—	—	—	—	—	1	4
Total	1	105	9	4	1	2	—	—	—	—	1	-	1	124

is $14,8 \pm 8,0$ and D/m = $1,85$, the odds of this difference being significant are only 16 to 1. Thus, although minor differences in the degree of constancy may perhaps occur, the main result is the remarkable constancy and the almost complete inability of the mother plants to produce offspring with a lower chromosome number than 22. This result was, indeed, quite unexpected, since mother plants with 25 and 24 chromosomes had been found to produce a great proportion of daughter plants with lower chromosome numbers than the mothers.

When looking for an explanation of the cytological constancy, the first possibility is of course the suggestion that the mother plants had become apomictic. In such a case there should have been a sudden and simultaneous change from sexuality to apomixis, affecting all the nine mother plants, or at least the seven individuals that were picked out directly from the purely sexual material. This does not seem very

probable, indeed, and the morphological and cytological results obtained suggest another explanation.

As always in *Poa alpina,* under the present conditions of cultivation, morphological observations were sometimes hampered by a rather high mortality, many plants having died before the morphological notes could be made. Nevertheless, it was obvious that the progenies under discussion, having predominantly 2n = 22, were not quite as uniform as apomictic progenies cultivated at the same time. In the first place this was evident by a greater variation in vigour. In 1937 vigour was estimated in a number of progenies of plants with 2n = 22, and at the same time comparable material of some apomictic types was available. The following values were obtained (Table 7).

TABLE 7. *Vigour in some apomictic alpina strains and in a sexual strain with 22 chromosomes.*

Progeny No.	Vigour							n	M ± m	σ^2	v
	0	1	2	3	4	5	6				
1935--1 (apomictic)	2	—	3	2	24	14		45	3,96 ± 0,17	1,36	29,5
--2 »			1	1	30	20	8	60	4,55 ± 0,11	0,67	18,0
—3 »	2	—	4	15	22	7	3	53	3,66 ± 0,17	1,46	33,1
—4 »	2	—	—	5	16	7	4	34	3,94 ± 0,23	1,82	34,3
1935—14 (sexual, 2n = 22)	4	3	2	4	13	9	1	36	3,39 ± 0,29	2,94	50,7
-16 » »	5	2	6	2	8	6	1	30	2,93 ± 0,34	3,45	63,5
—18 » »	3	2	—	3	11	8	3	30	3,77 ± 0,32	3,02	46,2

A glance at the distribution will at once show that the last three series in the table (»2n = 22») are more variable than the apomictic progenies. This is confirmed by a calculation of variances and coefficients of variation. Among the apomictic progenies the variance values (σ^2) range from 0,67 to 1,82 and the v-values from 18,0 to 34,3. Among the progenies from mother plants with 2n = 22 the variance values range from 2,94 to 3,45 and the v-values from 46,2 to 63,5. Thus, it is quite clear that the last-mentioned progenies are more variable, in spite of the fact that cytologically they are just as stable as the apomictic progenies. Disregarding the low proportion of aberrants in both categories, the only possible explanation of the observed difference in variability must be the assumption that in the progenies with 22 chromosomes recombination is still going on in contrast to the

apomictic progenies, in which all individuals have the same genotypical constitution.

3. MEIOTIC OBSERVATIONS.

A. REGULAR MEIOSIS IN A PLANT WITH 2n = 22.

The assumption of a genotypical recombination in strains with a stable chromosome number of 22 seems strange when considering the high degree of chromosomal variation in all related sexual progenies and the high degree of meiotic irregularities observed in two of the apomictic biotypes. Nevertheless, cytological observations in one of the mother plants with 2n = 22 confirmed the assumption. In plant No. 57 (cf. Table 6) *first metaphase was found to be characterized by a regular occurrence of 11 bivalents.* Two complete I—M groups from this plant are represented in Figs. 12 and 13, and in both these groups there is a perfect regularity. Many other I—M groups showed the same regularity, and in some of them the chromosome configuration was analysed and found to be 11_{II}. No univalents or multivalents were seen at this stage, and also interphase, second metaphase and anaphase were apparently quite free from disturbances. Only in one single case was an eliminated chromosome observed at II—M.

The observed meiotic regularity evidently explains the constant chromosome number in the offspring. According to Table 6, 27 daughter plants have been raised from plant No. 57 and of these 25 were found to have 2n = 22. Two granddaughter plants (from plant No. 144) also had 22 chromosomes. — Though meiosis was only examined in one of the mother plants there is every reason to believe that the constant or almost constant chromosome number in the other progenies is due to a similar meiotic regularity in the respective mother plants. Thus, *from a material with oscillating chromosome number a stable strain has arisen,* in which chromosome variation is very slight. It is quite remarkable that this constancy should be reached at the chromosome number 22, the basic number of the genus *Poa* undoubtedly being 7 (cf. MÜNTZING, 1932, p. 147 and NANNFELDT, 1937 a). This transition from the basic number 7 to the basic number 11 will be further considered below (p. 182).

B. IRREGULAR MEIOSIS IN A PLANT WITH 2n — 26.

That the regular 22-chromosome plants are exceptional among the sexual material studied is also evident from observations made on a

sexual plant with 2n = 26. This plant was one of the nine plants raised from the original seed sample (cf. MÜNTZING, 1932, p. 133). Meiosis in this individual was found to be quite irregular and of a similar type to that of the apomictic strains studied.

Thus, at I—M univalents and trivalents were observed in addition to bivalents. Probably also larger associations than trivalents occurred but in a low frequency. The following configurations were observed: $1_{III} + 11_{II} + 1_I$ (2 cells); $1_{III} + 10_{II} + 3_I$ (1 cell); $2_{III} + 9_{II} + 2_I$ (1 cell) and $3_{III} + 8_{II} + 1_I$ (1 cell). The two last-mentioned configurations are probably correct, but owing to poor fixation they are not entirely reliable. The group with $1_{III} + 10_{II} + 1_I$, on the other hand, was quite clear.

At first anaphase division of univalents was observed and at interphase micronuclei were frequent. Eliminated chromosomes were also of a common occurrence at II—M, and at II—A lagging chromosomes were seen. The frequency observed was the following:

Number of lagging chromosomes: 0 1 2 3 4
»   » II—A groups: 24 20 18 5 2

The average number is 1,14, which corresponds well with the number of univalents observed at I—M.

Though the meiotic observations in this plant are fragmentary, they are evidently sufficient to show that a plant of this kind will form gametes with variable chromosome numbers, and that there is a certain degree of chromosome elimination. These observations are in accordance with the fact that in progenies of sexual *alpina* plants, with more than 22 chromosomes, the chromosome numbers were found to be variable and on an average lower than in the mother plant. Unfortunately, no progeny was raised from the particular plant studied, but progenies of some sister plants gave the chromosomal variation represented in Table 2. Under such circumstances it is clear that meiosis in these sister plants was of the same irregular type.

C. ON THE OCCURRENCE OF UNREDUCED POLLEN GRAINS.

High chromosome numbers among the sexual material were not only obtained by raising progenies from plants with at least 31 chromosomes. In one case such high numbers were also obtained in the progeny of a mother plant with only 24 chromosomes. This plant was isolated and gave a total of 22 seeds in the isolation bags. From these

seeds only three daughter plants were obtained. These plants were rather poor in vigour and to my surprise they were found to have the somatic chromosome numbers 45, 46 and 48 respectively. Since these plants represented an approximate or exact doubling of the chromosome number, the pollen of the mother plant was studied. The suspicion that the chromosome doubling was brought about by the union of unreduced gametes was confirmed by pollen measurements. At the same time it was found, however, that two classes of pollen grains were not always produced by the plant in question.

Pollen samples were measured three times. the samples being collected on June 10th, 13th and 16th (1933). The distributions found were the following:

	Pollen diameter													
Units:	15	18	21	24	27	30	33	36	39	42	45	48	51	n
June 10th:	1	9	19	53	14	21	23	9	9	3	3	1		165
» 13th:	18	25	71	41	12	13	3							183
» 16th:	1	2	16	30	11	34	49	15	3	2	2			165
Total:	20	36	106	124	37	68	75	24	12	5	5	1		513

In the first and especially the third sample the distribution is bimodal, one maximum being situated between 24 and 27, the other between 33 and 36. The sample taken on June 13th, on the contrary, does not show more than one maximum. Evidently this maximum corresponds to the lower maximum in the other two series. If all values obtained from the plant are added, the total series will be bimodal. It is highly probable that the lower maximum corresponds to reduced pollen grains, the higher maximum to unreduced grains, and thus the pollen measurements partly confirm the conclusion previously reached that the plant in question is capable of forming unreduced gametes on the male as well as on the female side. It is interesting that this capacity, judging from the measurements, is not always at work. Under certain environmental conditions only reduced pollen grains are formed.

III. NEW POA ALPINA MATERIAL FROM SWITZERLAND.

According to the results described above the Scandinavian strains of *Poa alpina* were found to be quite or almost quite apomictic, in contrast to the material obtained from Switzerland. This is especially true f the material from Fürstenalp, which, as far as can be judged, is

purely sexual. The strain from St. Gothard, however, was predominantly apomictic but less stable than the Scandinavian apomicts.

Since it seemed desirable to obtain more information about the possible existence of an average difference between the Scandinavian and Swiss *Poa alpina* types as regards the mode of reproduction, more material from Switzerland was procured. For this material I am greatly indebted to Doctors N. SYLVÉN and G. NILSSON-LEISSNER. During a journey in Switzerland in 1934 these gentlemen collected seed samples of *Poa alpina* as well as *pratensis*, care being taken to collect the seeds from single individuals. Thus, though the risk that the seeds in a sample were gathered from more than one ind'vidual is not quite excluded, most of the progenies raised are certainly derived from single mother plants. The seeds were germinated in August 1934, and besides new material of *Poa pratensis* this germination resulted in 8 new *Poa alpina* progenies. In addition to these progenies two viviparous clones of *Poa alpina* were raised from bulbils also brought to Svalöf by Doctors SYLVÉN and NILSSON-LEISSNER.

In the material thus obtained chromosome counts and morphological observations were undertaken in the following years. The main result of these studies is that *predominantly the new Swiss material is apomictic, but in some strains varying degrees of sexuality occur.* This conclusion is based on the following detailed evidence.

1. CONSTANT OR APPROXIMATELY CONSTANT PROGENIES WITH $2n = 37$.

The chromosome number 37, previously met with in the strain from St. Gothard (cf. above p. 116), recurred again in several of the

TABLE 8. *Chromosome counts in Swiss Poa alpina strains, having*
$2n = 37$ as a typical chromosome number.

Field number	Origin	Chromosome numbers									
		25	... 33	34	35	36	37	38	...	46	... 57
34—36.........	Arosa					1	23	1			
—37.........	»					1	19	4	—	—	—	1
—38.........	»			1	1	1	—	20	—	—	1	
—39.........	Hasliberg						24	1			
—48.........	Rigi	1	—	—	—	—	—	16	1		
—50.........	Oberalp				1	1	4	15	5		

new Swiss strains. Chromosome counts in this group of the material gave the results represented in Table 8.

In the progenies 34—36 and 34—39 the chromosome number is probably quite constant. The three apparent aberrants, in which the counts had given the values 36 and 38, were morphologically quite typical, and therefore the deviations in chromosome number are probably due to slight errors in counting. The progenies in question were not only uniform in chromosome number but also in morphology. Thus, the conclusion must be drawn that in these biotypes apomictic seed formation is as regular as in the Scandinavian *alpina* apomicts.

Two other biotypes were found to show about the same constancy, viz. 34—37 and 34—48, but in these strains there were a few clear aberrants. In 34—37 the plants with 36 and 38 are no sure aberrants, but there was one plant having $2n = 57$. This plant is evidently the result of the union of one unreduced and one reduced gamete and is analogous to similar aberrants observed in some of the Scandinavian apomicts. This plant was somewhat weak, but otherwise it did not differ conspicuously from the other plants in the progeny.

In progeny 34—48 one plant was found to have $2n = 25$ but had not been observed to differ morphologically from the other plants of the progeny. In this case an experimental error does not seem to be altogether excluded. At any rate this biotype is characterized by a high degree of apomictic seed formation, 37 being the typical chromosome number.

In the remaining two progenies of the 37-group, however, a certain proportion of true aberrants were present. In progeny 34—38, 20 plants had the typical number 37, the chromosome numbers in 3 other plants being 33, 34 and 35 respectively. These plants, especially the one with 33 chromosomes, were observed to be clearly deviating morphologically already before the chromosome numbers were known.

In this family is seemed desirable to study the offspring of the apparent aberrants, and therefore progeny was raised from open-pollinated mother plants. In the four families studied the chromosome numbers were found to vary as follows:

Field No.	Chromosome number of the mother plant	Chromosome numbers in the offspring							
		32	33	34	35	36	37	38	39
1938—27	37						7	1	2
—28	35		1	7					
—29	± 34		2	---	4	1			
—31	33	2	7	1					

These values conclusively demonstrate that the strain under consideration, 34—38, is partially sexual and that the three plants found to have 33—35 chromosomes were true aberrants.

A quite similar situation was met with in strain No. 34—50 (Table 8). Of the apparent aberrants the one with $2n = 34$ had died at an early stage and the one with $2n = 35$ was observed to be slightly deviating in appearance before the chromosome number was known. As regards the other aberrants with 36 and 38 chromosomes, it is uncertain whether they represent true deviations from the typical chromosome number 37.

Since all the strains hitherto discussed have 37 as their typical chromosome number, they should perhaps be expected to be morphologically identical or similar. With one exception, however, all the strains were found to be dissimilar. The three strains from Arosa were clearly different but, curiously enough, one of the Arosa strains, 34—36, was found to be indistinguishable from the Rigi strain, 34—58. The type from Oberalp, 34—50, was also found to be very similar to the two strains just mentioned, but it differed rather clearly in some minor respects, the leaves being darker green and the spikelets more brownish red. It seems certain, however, that this strain must be genotypically quite closely related to the other two.

2. A MORPHOLOGICALLY HETEROGENOUS PROGENY WITH $2n = 33$ AS THE TYPICAL CHROMOSOME NUMBER.

A fourth strain from Arosa, 34—44, differed from the preceding three strains (with $2n = 37$) by having $2n = 33$ as the typical chromosome number. Chromosome variation in the initial progeny was found to be the following:

Chromosome number:	32	33	34	...	49
Number of individuals:	1	21	1	...	1

There is only one clear aberrant, viz. the plant having $2n = 49$. As probably all the other plants have 33 chromosomes, the morphological appearance of this strain might be expected to be quite uniform. Strangely enough, this was by no means the case, the family on the contrary being characterized by a marked morphological variation. With some difficulty, however, most of the plants could be divided into two morphological groups. This suggested the possibility that the material might be composed of a mixture of two different apomicts,

having the same chromosome number. In order to study this rather peculiar and interesting material more closely, progenies after isolation were raised from 6 morphologically dissimilar plants. The morphological properties of these progenies have not yet been studied, but chromosome counts are available. The deviating plant with $2n = 49$ gave 3 daughter plants having the chromosome numbers 49, 72 and 74 respectively. Evidently the last two plants resulted from the union of unreduced and reduced gametes. The other five plants in the original progeny, having $2n = 33$ or possibly $2n = 34$, gave rise to daughter plants showing the following chromosome numbers:

Field number	Chromosome number of the mother	Chromosome numbers in the progeny				
		31	32	33	34 ... 49	
1938—32	34 (33?)			4	1	
—33	33		3	6	— ... 1	
—34	33			10		
—35	± 33		1	4		
—36	33	1	—	8	1	

Evidently, most of the plants have $2n = 33$, and it is as yet uncertain whether the numbers slightly higher or lower than 33 represent aberrations or slightly erroneous counts. The production in progeny 1938—33 of a plant with $2n = 49$ is interesting, since such an individual, with 50 per cent higher chromosome number than usual, was also observed in the previous generations. The latter plant gave rise to individuals with $2n = 72$ and 74 and, thus, through the functioning of unreduced gametes in two consecutive generations,, individuals having the number 33 may rapidly give rise to products with more than seventy chromosomes.

3. A PROGENY COMPRISING MANY DIFFERENT CHROMOSOME NUMBERS.

The highest degree of chromosomal variation was met with in the progeny 34—46, in which the chromosome counts gave the following result:

Chromosome number:	26	27	28	29	30	31	32	33	34	35	36	37...52
Number of individuals:	10	2	—	—	1	—	1	2	2	--	—	1 ... 3

Eight different numbers were met with, of which 26 was the most frequent. Corresponding to the chromosomal variation the progeny

was also heterogenous morphologically. With some difficulty five different morphological types could be distinguished. Fifteen plants belonged to the first morphological type, and of these 15 plants 12 had been cytologically examined. These plants were all found to have either 26 or $\pm$ 27 chromosomes. The second morphological type comprised 5 plants, which were afterwards found to have 32, 33, 33, 34 and 34 chromosomes respectively. The third type comprised one plant having $2n = 37$ and the fourth type one plant having $2n = 30$. Finally, the fifth morphological type comprised the three plants having $2n = 52$. This type was especially conspicuous by broad, thick leaves and few and coarse panicles. — Thus, in this progeny there is evidently a rather good correlation between different chromosome numbers and different morphological types.

In order to get a deeper understanding of the cytological and morphological variation in this material, nine plants of the original family were isolated and progenies raised. The results of some chromosome counts from this material are available and also morphological data from two of the progenies, which were raised earlier than the others. These two progenies were both derived from mother plants with $2n = 26$. In the offspring the following numbers were observed:

Chromosome number:	24	25	26	27
Progeny 1:			6	
» 2:	1	—	3	1

Twenty-six is evidently the typical number, and it is uncertain whether the two deviating numbers observed represent true aberrants. Morphologically, progeny 2 seemed to be quite uniform, in contrast to progeny 1. This progeny consisted of 12 plants, of which 9 were typical in appearance, the remaining three representing a special deviating morphological type. The chromosome number 26 was observed in both the types, viz. in 4 typical and 2 deviating plants.

Four other progenies were raised from mother plants with chromosome numbers ranging from 30 to 37. In the offspring the following chromosome numbers were observed:

Progeny	Chromosome number of the mother	Chromosome numbers in the offspring						
		31	32	33	34	35	36	37 ... 43
3	30	1	1	8				
4	33		1	7	1			
5	34			9	1			
6	37			1	—	—	—	8 ... 1

In progenies 3, 4 and 5, 33 seems to be the typical chromosome number, and therefore it is somewhat doubtful whether the mother plants really differed in chromosome number. It should be remembered, however, that the mother plant of progeny 3 differed morphologically from the mother plants of progenies 4 and 5, the latter two being of the same morphological type. Progeny 6, finally, shows that the chromosome number of the mother plant was correctly determined and that this plant must have been predominantly apomictic although partially sexual. There are two clear aberrants among a total of 10 plants.

The last three progenies of the family under discussion (34—46) were raised from the three plants having $2n = 52$. Most of the daughter plants had the same or approximately the same number as the mother plants, but in addition to these a haploid plant appeared. The chromosome counts in the different progenies gave the following result:

Progeny 7: 54, 52, 52 and ± 27
        »   8: 51, ± 51, ± 52
        »   9: 51, ± 51, 52, ± 52, ± 52, ± 53

The appearance of the individual with $2n = \pm 27$ is quite interesting, since it suggests a direct relationship between the number 26 and 52, observed in the original progeny. Probably the change between 26—52 may go in both directions, and sometimes the union of unreduced and reduced gametes in individuals with $2n = 26$ may also give rise to products with intermediate numbers. This seems to be the best explanation of the chromosomal variation observed in the progeny 34—46.

4. TWO VIVIPAROUS CLONES.

As already mentioned above, the new material from Switzerland also included two viviparous clones, in which no seed production at all could be observed. Both these clones were collected in Arosa, but were found to be clearly different in morphology and were also found to differ in chromosome number. In one of them the chromosome numbers of 10 plants were examined, the result being $2n = 33$ in 9 plants and ± 34 in one individual. The other clone was characterized by the chromosome number 26, this number being counted in 7 plants.

These viviparous clones might perhaps be expected to show quite new chromosome numbers, but rather interestingly that was not the case. The number 33 was met with in several Scandinavian and also

in some Swiss agamospermic *alpina* strains, and the number 26 was found to be the most frequent number in the family 34—46, discussed above. It is interesting to note that this strain, as well as a strain with $2n = 33$ (34—44), were also collected in Arosa, the place from which the viviparous biotypes were obtained. This certainly suggests a relationship between the agamospermic and viviparous biotypes having the same aneuploid chromosome number.

5. CONCLUSIONS.

The new material from Switzerland was originally expected to be purely sexual, like the old Swiss material from Fürstenalp. Evidently, however, the degree of apomixis in the new strains is on an average rather pronounced, and not a single strain was found to be quite sexual. In one strain, however, the percentage of aberrants seems to be quite high. In other strains there is a low but quite clear proportion of aberrants, and in still other strains apomixis seems to be absolute. The most frequent deviations in chromosome number were due to the functioning of unreduced gametes, generally resulting in the union of one unreduced and one reduced gamete or sometimes also in the union of two unreduced gametes. In some cases aberrants were also formed, presumably by the union of two reduced gametes. The functioning of unreduced gametes will tend to increase the chromosome number, but the opposite process, the formation of haploids from types with high chromosome numbers, was also observed. Judging from chromosome numbers, viviparous and agamospermic strains from the same locality may be rather closely related.

IV. CROSSES BETWEEN SEXUAL AND APOMICTIC STRAINS.

Since in our *Poa alpina* material the occurrence of strains with apomictic seed formation as well as strains which apparently are quite sexual had been established, it was natural to carry out hybridization experiments between representatives of the two categories. By producing hybrids between sexual and apomictic *alpina* plants and by studies of their offspring some information about the genetic basis of apomixis in this material might be gathered. Crosses of this kind were performed in 1933 and since then F_1-, F_2- and to some extent also F_3-generations have been studied with the following results.

1   THE PARENTAL PLANTS.

Two sexual mother plants are involved in the crosses, 32—53—44
and 32—53—16   Both plants have 2n = 24 and were members of the
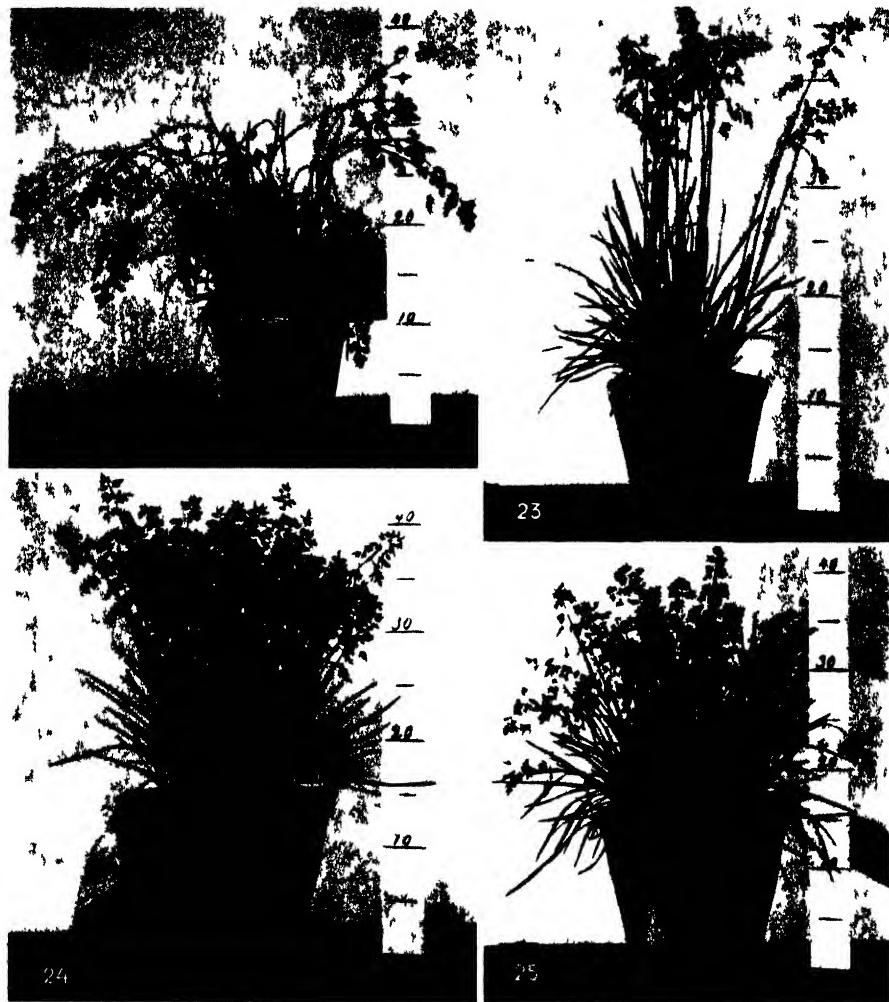
Figs 22—25   Parental plants and two F_1 hybrids from the cross sexual × apomictic
Poa alpina — Fig 22, a sexual mother plant (2n = 24), Fig 23, the apomictic
pollen parent (2n = 38), Fig 24, an F_1 plant with 2n = 27, Fig 25, an F_1 plant
with 2n = 41 (resulting from the union of an unreduced ovule and a reduced
male gamete)

progeny 32—53, the chromosomal variation of which is given in Table 2.
The first plant, 32—53—44, had dehiscing anthers and rather good

pollen, the percentage of good pollen in three samples taken being 87, 78 and 84. The first value is from a sample taken in 1933, the latter two values being obtained in 1935. The other sexual plant, 32—53—16, is fertile on the female side, but in 1933, when the crosses were made, it was found to be male sterile, having non-dehiscing anthers. A later observation, in 1935, showed, however, that this male sterility is not absolute, two pollen samples obtained in this year showing 78 and 84 per cent good pollen.

Morphologically (cf. Fig. 22), the two sexual mother plants do not present any specially striking characteristics except the fact that in both of them, as in all other members of the strain from Fürstenalp, the spikelets are purely green, being quite free from anthocyanin.

The father plants used all belonged to the Korpilombolo apomict from North Sweden (Fig. 23). This strain has $2n = 38$ and is characterized by a very good pollen fertility (p. 120, strain 1), although the chromosome number is aneuploid, and meiosis was found to be irregular (p. 125). In this strain the spikelets were not green but coloured by anthocyanin. The father plants were taken from this strain on account of its good pollen and relatively high chromosome number. In order to decide whether the crosses undertaken had been successful, it seemed desirable that the difference in chromosome number between the parent plants should be relatively great.

When making the crosses, in 1933, the parent plants were simply put together in pairs in isolation cages, placed in a greenhouse. No attempts were made to emasculate the flowers of the mother plants. In the case of the male sterile mother this was superfluous, but also from the other mother plant most of the seeds obtained gave rise to true F_1 hybrids.

2. THE F_1-FAMILIES.

A. CHROMOSOMAL VARIATION.

After the cross-pollinations about 500 seeds were obtained from the mother plant 32—53—44 and 73 seeds from the other female parent. These seeds were germinated in the same summer, about a month after the harvest, and gave rise to 289 and 34 seedlings respectively. During the following winter many plants died, but in the summer of 1934 a total of 214 plants were still available for observation. Root tips of the young plants had been fixed and this enabled determinations of chromosome number in a total of 202 plants.

All the daughter plants of the male sterile mother had anthocyanin-

coloured panicles like their father and, thus, they were true F_1 hybrids. This was also evident from their chromosome numbers. — In the offspring of the other sexual mother plant the great majority of the individuals were F_1 hybrids, showing the anthocyanin colour and having more or less high chromosome numbers. Only 3 plants were observed to be the result of self-pollination, these plants having green panicles and relatively low chromosome numbers (24, 25 and 27).

As it seemed desirable to study the chromosomal variation in F_1 as closely as possible, the chromosome numbers of as many as 202 F_1 plants were determined. The result was the following:

Chromosome numbers in F_1

F_1-progeny No.		25	26	27	28	29	30	31	32	33	34	35	. . .	40	41	42	43	n
1	:				5	6	4	5	6	1	—	—	. . .	1				28
2	:	1	1	7	17	34	38	31	27	10	2	1	. . .	—	3	1	1	174
Total	:	1	1	7	22	40	42	36	33	11	2	1	. . .	1	3	1	1	202

Progeny 1 in the above table was derived from the plant 32—53—16, progeny 2 from 32—53—44. In the latter progeny the chromosome numbers of the non-hybrid plants have been excluded. Thus, in the table only chromosome numbers of true F_1 hybrids are included.

The chromosomal variation in F_1 is evidently bimodal, one maximum being situated at 30, approximately, the other one at 41. The cause of this bimodality is obvious. *The lower maximum corresponds to the union of reduced male and female gametes, the higher maximum to the union of reduced male and unreduced female gametes.* Theoretically, the former maximum should be situated at 31, this value being the sum of 12 and 19. The second maximum should have the value 43 (24 + 19). The slight decrease in the values observed is most probably due to a certain extent of chromosome elimination at meiosis. Such elimination was observed in the male biotype and may also be expected to occur in the female plant.

The wide variation in chromosome number demonstrates that meiosis must be irregular at least in one of the parents. The following facts demonstrate that not only the male but also the female parents are responsible for this variation. Firstly, the mother plants belong to a material characterized by its oscillating chromosome number. Secondly, the 3 plants obtained by self-fertilization of one of the mother plants were found to represent three different chromosome numbers (24, 25 and 27, as mentioned above). On another occasion the same individual was isolated and gave rise to three daughter plants with 2n = 24, 25 and 25 respectively.

B. VIGOUR.

The 214 F_1 plants available for observation in 1934 represented all degrees of viability, ranging from dying to very vigorous plants (cf. Figs. 24—25). On an average vigour was found to be higher in F_1 than in the sexual parent strain. In 1934 this strain was represented in the field by 88 individuals, which could be compared with 164 F_1 plants. The remaining F_1 plants were grown in a greenhouse. Using the usual scale from 0 to 10, vigour in this material was estimated with the following result:

	Vigour						
	0 —	2 —	4 --	6 —	8 —	10	n	M
Sexual mother strain:	35	21	5	21	6		88	3,68
F_1-plants:	16	51	39	31	27		164	5,02

Owing to the skewness and bimodality of the first series the standard errors were not calculated. Nevertheless, it is rather clear that the average of F_1 is better than that of the mother strain. The number of plants available of the paternal biotype was not sufficient to allow exact measurements. From later observations, however, it is rather clear that the average vigour of F_1 is somewhat lower than that of the male parent, but some F_1 plants are even more vigorous than the male parent plants.

Since the F_1 plants represent many different degrees of vigour as well as chromosome number, an attempt was made to find a correlation between chromosome number and vigour. But again the result was quite negative. The different vigour classes had about the same average chromosome number. Though it does not seem necessary to publish the whole correlation table, the absence of a correlation may be demonstrated by the following average values:

Vigour classes	: 0 —	2 —	4 —	6 —	8 —	10
Average chromosome number:	31,1	30,5	30,6	30,7	30,7	
Number of individuals:	16	51	39	31	27	

The chromosome numbers of the F_1 plants involved in this correlation ranged from 25 to 35. The individuals having $2n = 40$—43 were all placed in pots in the greenhouse, and thus they were not comparable to the field plants. Their viability was good, however, as may be seen from Fig. 25.

C. FERTILITY.

As the F_1 hybrids were obtained from crosses between *Poa alpina* strains of widely different origin it seemed desirable to test whether the F_1 plants showed any signs to hybrid sterility. Studies on pollen fertility demonstrates, however, that the F_1 plants are comparable in this respect to the sexual mother strain. The following values were obtained:

| | Per cent good pollen | | | | | | | | |
	40 — 50 — 60 — 70 — 80 — 90 — 100							n	M
Apomictic parent				1	7	20		28	91,8
Sexual »		1	1	10	9			21	87,9
F_1 hybrids	3	2	7	17	39	35		103	83,6

The F_1 hybrids have the lowest average value, but the difference between the values of F_1 and the sexual parent strain is certainly not significant. In addition to the plants giving pollen samples male sterile individuals with non-dehiscing anthers were observed in F_1 as well as in the sexual parent strain, but their exact number is not known.

Though, in analogy to previous results in the sexual parent strain (cf. above, p. 129), fertility might also be expected to be independent of chromosome number, it seemed desirable to test the existence of such a correlation in the present F_1 material. This test gave the following values:

| Per cent good pollen | Chromosome number | | | | | | | | | n | M |
	26 — 28 — 30 — 32 -- 34				36	38 — 40 — 42					
Lower than 80: ..	1	8	11	7						27	30,8
80— 90:	2	14	16	7						39	30,4
90—100:	2	6	12	7	1	—	- -	2		30	31,6

The three pollen fertility classes evidently contain plants, having about the same average chromosome number, and thus no correlation can be established. Though negative, this result is rather interesting.

Seed setting in F_1 was not studied in detail, but it seems to be rather good. 50 F_1 plants were isolated in various ways, and in 45 of these plants various degrees of seed setting were obtained, ranging from 1 to more than 100 seeds per panicle.

D. MEIOSIS.

Studies of meiosis in F_1 are limited to some observations in an F_1 plant having 2n = 41, and thus belonging to the group of F_1 individuals arisen from the union of unreduced ovules and reduced male gametes.

The plant studied is the mother plant of the progeny 34—19 (Table 9). Meiosis in this plant was found to be somewhat irregular but without profound disturbances. At I—A division of a few univalents was often observed (Fig. 11). At interphase one or two micronuclei were frequently seen, and there were often some eliminated chromosomes outside the II—M groups. The I—M groups were characterized by a frequent occurrence of multivalents. In Fig. 9 a number of such multivalents are represented together with some bivalents and a univalent. The bivalents and the univalent are from the same I—M group as the large multivalent, which is probably composed of seven chromosomes. The other multivalents probably represent one quinquevalent, two quadrivalents and one trivalent. Fig. 10, finally, represents a I—A with the distribution 20—21.

3. F_2-PROGENIES.

Since the cross was made in order to study the inheritance of apomixis, the first question to be answered is whether sexuality or apomixis is dominant. If the F_1 plants are apomictic, like the pollen parent, they should breed true, in the opposite case they should segregate. In the present material the question may be rather easily answered owing to the variable and aneuploid chromosome numbers. If a plant breeds true, the chromosome number in the offspring will remain constant, in the opposite case segregation will practically always be accompanied by a variation in chromosome number.

Progenies were raised after isolation from 17 different F_1 plants, care being taken to select, in the first place, F_1 plants with odd chromosome numbers. Thus, most of the F_1 plants involved had 27, 29, 31 or 33 chromosomes. Progenies were also taken from some F_1 plants with $2n = 41$. The results of chromosome counts in these progenies are given in Table 9.

A glance at the table is sufficient to show that *without exception the F_2 progenies had variable chromosome numbers*. Since this variation is very marked, there is reason to believe that the mother plants are purely sexual. These mother plants were taken at random (apart from the fact that most of them had odd chromosome numbers) and therefore they may be regarded as representative of the entire F_1 generation. Thus, it may be concluded that *in this material apomictic seed formation is recessive to sexual propagation.*

Considering the type of chromosomal variation, the functioning of unreduced gametes has evidently occurred in some progenies. Thus,

TABLE 9. *Sexual × apomictic Poa alpina. Chromosomal variation in F₂.*

Field number	— Somatic chromosome numbers																										n	M	Chromosome number of the mother	Difference
	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44				
34—13						2	1	4	5	2	3	2										1		1			28	28,2	27	+ 1,2
—14						1		2	2	2	1	3	1		1												11	29,8	29	+ 0,8
—15							1	1		3	2	1	1														11	30,6	31	— 0,4
—16											1	1	1														3	32,0	33	— 1,0
—17			1	3	2	1	1	1										1		8	14	9	2	2		1	54	37,0	41	— 4,0
—18																			1	8	8	2	2	1			7	39,4	± 41	— 1,6
—19		1								1	1		1								2	2	1				6	37,5	41	— 3,5
—21	1							1	1	3	2		1								1	2	1	1			8	28,3	29	— 0,7
—22							1		1	1																	3	28,0	27	+ 1,0
—23								1		1	3					1											5	32,6	31	+ 1,6
—24											1	1		1			1										2	31,0	31	0,0
—25									1	1	1	2	3	1		1											10	32,4	33	— 0,8
—26								2	1	6	7	5	2	1	3	3											24	29,9	31	— 1,1
—27								1	2	3	3	2															11	29,3	29	+ 0,3
—28							1		1	2	2	1				1											7	29,1	29	+ 0,1
—29								3	1	3	4	4	1	1	1												18	30,6	30	+ 0,6
—30						1				2	2	2	1	1		1							1				9	31,8	28	+ 2,8

in the first progeny, 34—13, there are two exceptional plants having 41 and 43 chromosomes. Since the chromosome number of the mother plant is 27, these aberrant plants are evidently the result of unreduced + reduced gametes. The same story is repeated again in progenies 34—29 and 34—30 with respect to the aberrant plants having 2n = 44 and 40 respectively, the chromosome numbers of the mothers in this case being 30 and 28.

This functioning of unreduced gametes has already been described above for several other *Poa alpina* plants. Of more interest than this phenomenon is the fact that *the mother plants with the highest chrosome numbers* (2n = 41) *are capable of forming a considerable proportion of haploids.* Thus, in the progeny of this kind most extensively studied, 34—17, not less than 8 plants were found to be haploids in relation to the mother plant, their chromosome numbers ranging from 19 to 23. *The percentage of spontaneous haploids in this progeny is as high as 14,8 per cent.* Since a haploid also appeared in the analogous progeny 34—19, it is probable that this spontaneous formation of haploids is characteristic of all these F_1 plants, which resulted from the union of reduced and unreduced gametes. A similar production of a plant with approximately half the chromosome number of the mother plant was also observed in one of the progenies of the new Swiss material. In this case a mother plant with 2n = 52, which probably was the result of doubling of the typical chromosome number 26, produced a haploid having 2n = ± 27 (cf. p. 144).

The great majority of the F_2 plants had chromosome numbers varying around the values of the corresponding F_1 plants. In the sexual parent strain there was a rather clear tendency for the progenies to have slightly lower average numbers than the mother plants (cf. pp. 128 and 131) and this tendency was ascribed to chromosome elimination at meiosis. In the present material, however, this tendency seems to be less marked, and a calculation of the average values of the progenies shows, in fact, that it is quite absent. As is evident from Table 9, 8 progenies have lower average chromosome numbers than the corresponding F_1 plants, 8 progenies have higher values and in one case the difference is ± 0. Thus, *the chromosome variation produced in F_1 by crossing types with 24 and 38 chromosomes, is retained in F_2 in a rather unchanged condition.* If progenies had been raised from all F_1 plants, the result would most probably have been the same. This absence of any chromosomal selection, combined with the negative results of all previous attempts to find a correlation between chromosome number,

vigour and fertility in *Poa alpina,* makes it unnecessary to look for correlations of this kind in the present F_2 material. Only the most extreme contrasts in chromosome number may be compared, viz. the haploids in progeny 34—17 and the corresponding normal plants. No accurate data on fertility are available but notes on the vigour of the plants have been made. These notes show that the haploids had the same average vigour as the other plants. The vigour distribution was the following:

	Vigour							
	0 — 2 — 4	6	–	8 — 10	n	M
The whole material:	11	25	46	13	2	97	4,38
The haploids:	1	2	4	1		8	4,25

Since these haploids did not differ significantly in vigour from the sister plants having twice as many chromosomes, there is certainly no correlation between chromosome number and vigour in the other progenies either, in which chromosome variation is less extreme. Nevertheless, all these progenies are characterized by a most pronounced variation in vigour as well as morphology, and in these respects they are markedly different from the apomictic strains. Undoubtedly, this increased variability is correlated with the oscillatory chromosome number, in spite of the fact that no average correlation could be found between chromosome number and vigour. Evidently, some constellations of chromosome and genes are more successful than others but these successful combinations may sometimes represent high and sometimes low chromosome numbers in the same manner as the unsuccessful combinations.

4. F_3-PROGENIES.

The study of the F_2 plants demonstrated that the F_1 mother plants were all sexual. Using the same chromosome counting method, it is necessary to raise a large F_3 generation in order to test to what extent the F_2 individuals are sexual or apomictic. In the simplest case, if the apomictic mode of seed formation were due to a single recessive gene, it should not be difficult to find F_2 plants giving constant progenies. Obviously the method used is very laborious, when it is necessary to study a large number of F_3 progenies, and as yet the work is far from having been accomplished. However, at the time of writing chromosome numbers of 154 F_3 individuals are available, these individuals belonging to 31 different families. Among these families not a single one has

proved to be constant. In 9 families the number of individuals is as yet too low to allow quite safe conclusions, but in the other 22 families there is a quite clear chromosomal variation, demonstrating that the mother plants are sexual or partially sexual. This is probably true also of the mother plants of the 9 families first mentioned. Under such circumstances *the simplest way of inheritance of apomixis seems to be quite excluded, viz. the possibility of apomixis being conditioned by a single recessive gene.* So far F_3 seems to behave in exactly the same way as F_2, indicating that all the mother plants in F_2 were just as sexual as the F_1 plants.

More definite conclusions may perhaps be drawn later on when more progenies have been studied, and when the chromosome counts have been supplemented with morphological and embryological data.

V. SOME NEW RESULTS IN POA PRATENSIS.

1. A SEXUAL HAPLOID.

A. INTRODUCTION.

In my previous paper on *Poa* (MÜNTZING, 1932) the occurrence of a *Poa pratensis* type with $2n = 36$ was reported (l. c. p. 145). The tuft containing roots with that number was not pure, however, many roots showing the double number $2n = 72$. The most plausible explanation seemed to be that the tuft consisted of two individuals growing together. This was found to be true by dividing part of the tuft into separate tillers. Some of these tillers were found to have broad leaves, others had narrow leaves. Ten tillers of each kind were planted in pots and the chromosome numbers were determined. Without exception the chromosome numbers in the broad-leafed tillers were found to be 72 or ± 72, all the narrow-leafed ones having $2n = 36$. These plants were then transplanted into the field for further observations.

B. FERTILITY.

Fertility in the haploid was found to be less good than in the corresponding type with 72 chromosomes. In the pollen the following values were obtained:

	Per cent apparently good pollen										
	55	60	65	70	75	80	85	90	95	100	n	M
Type with $2n = 72$							1	4	2		7	93,2
» » $2n = 36$	2	4	2	—	1						9	64,2

Though the number of pollen samples studied is small, it is obvious that the percentage of good pollen is much lower in the haploid than in the corresponding type with the double chromosome number.

The difference in fertility was found to be still more pronounced on the female side. As a rough measure of seed setting the number of seeds per panicle after open pollination was counted. In the 36-chromosome type a total of 389 seeds were harvested from 59 panicles, which corresponds to an average of $6,6$ seeds per panicle. In the corresponding 72-chromosome type an average of $432,1$ seeds per panicle was obtained, 7345 seeds being harvested from 17 panicles. The values in the two kinds of material are of entirely different orders, the average seed production in the 72-chromosome type being 65 times as high as in the type with 36 chromosomes.

For comparison seed production was counted in the same way in 9 other *pratensis* biotypes, having the ordinary high chromosome numbers and several of them being definitely known to be apomictic. The average value in this material was $458,4$ seeds per panicle. This value was obtained from counts of 23 panicles, the numbers of seeds per panicle varying between 201 and 692. Evidently, then, the 72-chromosome type is to be regarded as normal and the 36-chromosome type as exceptional.

C. MEIOSIS.

Some meiotic observations were made in the haploid. Since meiosis in other *pratensis* biotypes studied was found to be irregular (MÜNTZING, 1932, pp. 143—145; RANCKEN, 1934), and since the basic chromosome number in *Poa* is 7, meiosis in the rather sterile type with 36 chromosomes might, indeed, be expected to be quite irregular. To our great surprise, however, *meiosis in the haploid was found to be almost perfectly regular*, the typical chromosome configuration at I—M being 18_{II}. This configuration could be distinguished in 8 cells, no other type of association being observed. In Figs. 26—27 two typical I—M groups with 18_{II} are represented. Most of the bivalents seem to be ring-shaped, having at least 2 chiasmata. In rare cases multivalents may occur. A trivalent and a quadrivalent from different cells are represented in Fig. 31:

The rather high degree of regularity is also evident from the following counts of the number of univalents present at I—M.

Number of univalents at I—M: 0   1   2   3   n
»   » cells: 114   20   13   3   150

Thus of 150 cells observed 114 (76 per cent) were perfectly regular, 1 to 3 univalents being present in the other cells. Fig. 28 shows a I—M group with 2 univalents, Fig. 29 a rather exceptional cell with 8 uni-

Figs. 26—33. Meiosis in a sexual *Poa pratensis* biotype with 36 chromosomes. — Fig. 26, I—M side view (separately drawn), 18_{II}; Fig. 27, polar view, 18_{II}; Figs. 28—29, I—M groups showing different degrees of non-conjunction; Fig. 30, I—A, distribution 18—18; Fig. 31, one quadrivalent and one trivalent; Fig. 32, I—A, division of two univalents; Fig. 33, I—A, dicentric chromatid and fragment.

valents. (This cell was observed after the counts in the 150 cells were finished.)

At I—A the distribution 18—18 (Fig. 30) was counted in 8 cases. At the same stage the number of dividing univalents was counted in

100 cells. The result was 0 univalents in 95 cells, 1 univalent in 3 cells and 2 univalents in 2 cells. At interphase 97 p. m. c. were observed to be regular, and micronuclei were only seen in 3 cells. Fig. 32 shows a I—A with 2 dividing univalents and Fig. 33 a I—A with chromatin bridge and fragment. Thus in spite of the pronounced meiotic regularity, the individual in question must be heterozygous for an inversion or duplication.

D. PROGENIES AFTER OPEN POLLINATION AND ISOLATION.

Progenies of the biotype in question were raised after open pollination as well as isolation. Seed setting in the isolation bags was extremely poor, but nevertheless it was possible to raise some progeny. Chromosome counts were undertaken in a total of 54 daughter plants. Among these 25 individuals were raised from seeds of open-pollinated panicles, and 29 plants were the result of isolation. The former plants were *extremely variable in morphology as well as chromosome number, thus demonstrating that the mother plant is sexual.* Among the 25 plants the following 13 chromosome numbers were represented (the number of individuals in brackets): 32 (1), 39 (2), 42 (1), 43 (1), 44 (5), 45 (3), 48 (1), 52 (5), 53 (2), 54 (1), 55 (1), 56 (1) and 58 (1). At the time of flowering the haploid was surrounded by a collection of *pratensis* biotypes with chromosome numbers ranging from 49—82 (and possibly other more extreme numbers, since the chromosome number was not known in all biotypes). Evidently, these biotypes have been successful as male parents, not a single daughter plant having the same chromosome number (36) as the mother.

As already mentioned, the morphological appearance of the daughter plants was highly variable and they were all different from each other. It does not seem necessary to give any morphological descriptions.

In this material pollen fertility was also studied. Some plants were probably male sterile, no pollen being obtained. In the others the amount of good pollen was found to vary between 40 and 100, the average value being 77,0 per cent.

As regards chromosomal variation, the progenies obtained after isolation were found to be much regular. Of the 29 plants studied, 21 were found to have 2n = 36 like the mother plant. Of the 8 deviating numbers, 5 were close to 36 (one 34, three 35 and one 37), the others were much more aberrant (29, 49 and 52 respectively). The latter two values, 49 and 52, may be the result of unreduced ovules

fertilized by reduced male gametes. The main feature in this material, however, is not the aberrants but rather the high degree of constancy in chromosome number and the very marked contrast in this respect between the progenies after open pollination and isolation. Regarding the former, the haploid's own pollen was evidently unable to compete with the pollen of surrounding *pratensis* biotypes. On isolation, on the contrary, no other pollen was available, and a few seeds resulted from self-fertilization. Since meiosis was found to be predominantly regular, most gametes should have 18 chromosomes and this was, indeed, verified by the fact that most of the offspring had the same chromosome number as the mother.

For most of the material after isolation no morphological data are yet available. A few plants raised earlier than the rest, however, were found to be variable in appearance. This demonstrates once more that the constancy in chromosome number is not due to apomixis but to meiotic regularity. An exactly analogous case was described above (pp. 134—135), a sexual *Poa alpina* strain with $2n = 22$ being almost quite constant in chromosome number, due to a regular meiosis.

E. HYBRIDS WITH POA ALPINA.

The sexuality of the haploid *pratensis* type under consideration was also demonstrated by crosses with *Poa alpina*. In 1935 the haploid was pollinated with *alpina* pollen, two different *alpina* plants being used as the male parents. In the first cross 2 panicles pollinated gave only one seed, in the second cross 3 panicles were pollinated, and of these the first panicle gave no seed, the second 1 seed and the third 39 seeds. The seed obtained in the first cross gave rise to a pure *pratensis* plant having $2n = 35$. This plant was evidently the result of self-fertilization. The other 40 seeds resulted in 15 plants, which proved to be true *pratensis* $\times$ *alpina* hybrids. This was evident from their morphological characters as well as their chromosome numbers. Since *pratensis* $\times$ *alpina* hybrids have already been described by other workers (ÅKERBERG, 1936 b; NANNFELDT, 1937 b), it seems unnecessary to give a description of my own hybrids. Their chromosome numbers, however, are interesting and were found to have the following values:

	Chromosome numbers						
	31	32	33	34	35	...	52
Number of F_1 plants:	1	6	6	—	1	—	1

The father plant used for the crosses had $2n = 31$, and considering the probable occurrence of meiotic chromosome elimination most of

its gametes should have ± 15 chromosomes. Since most of the gametes of the mother plant have 18 chromosomes, the observed chromosome numbers of the F_1 plants (chiefly $2n = 32$ and 33) are in accordance with expectation. The individual having $2n = 52$ must result from the union of an unreduced ovule and a reduced male gamete. It demonstrates once more that functional, unreduced gametes are sometimes formed in the haploid.

F. PROGENIES OF THE SISTER TYPE WITH 72 CHROMOSOMES.

In the preceding section the type with 36-chromosomes was called a haploid and is supposed to be closely related to the type with the double chromosome number. The haploid is clearly sexual and almost sterile. Judging from the abundant seed production, the corresponding 72-chromosome type may be supposed to be apomictic like most other *pratensis* biotypes. In order to test that further, progenies after isolation as well as after open pollination were raised. Judging from morphological inspection alone, the biotype in question is predominantly apomictic but partially sexual, a certain proportion of deviating aberrants being formed. Pending the results of chromosome counts and further morphological observations, the exact degree of apomixis in this strain cannot yet be stated. At any rate the 72-chromosome type, however, must have a much stronger apomictic tendency than the 36-chromosome type, which seems to be purely sexual.

2. TWIN INVESTIGATIONS.

A. TRIPLOID AND HAPLOID TWINS.

In earlier publications (MÜNTZING, 1937, 1938 b) the production of plants with deviating chromosome numbers from twin seedlings was reported. The species studied also included *Poa pratensis*. As in the other species investigated, the most frequent deviation observed in *Poa pratensis* is represented by plants having their chromosome number increased by approximately 50 per cent. A total of twenty such »triploids» have been gathered. Morphologically it is rather difficult to distinguish »diploid» and »triploid» *pratensis* twins from each other, and even if the two twins are dissimilar in appearance, it is sometimes difficult to decide which of the two plants has the higher chromosome number. This difficulty is partly due to the fact that the twin with the higher chromosome number is more slow-growing and requires several years to reach full development (cf. below p. 162).

Another category of deviating twin plants may be easily distinguished also in *Poa pratensis*, viz. the haploids. So far only two diploid-haploid twin pairs have been observed in *Poa pratensis*, but in these pairs the haploids were definitely recognized as such by their smaller size and more slender mode of growth. The chromosome numbers in the first twin pair were found to be $\pm$ 72 and $\pm$ 36, and in the second pair $\pm$ 75 and $\pm$ 39 respectively. So far the properties of these haploid twins have not been studied, but their mere occurrence is of interest, especially with regard to the sexual haploid described above. Since, evidently, haploid twins may be formed, it is highly probable that the sexual *pratensis* type with 36 chromosomes arose as a twin from the original strain with 72 chromosomes. This haploid is rather vigorous, and it does not seem improbable that the original tuft, showing the mixture of tillers with 36 and 72 chromosomes, arose from one seed producing two viable seedlings with $2n = 36$ and 72. Gradually the tillers of these individuals became intermingled to such an extent that they could only be separated by division of the original tuft into small pieces.

B. THE PROPERTIES OF TRIPLOID TWINS.

a. *Plant weight.* — In order to study more closely the effect of a 50 per cent increase of the chromosome number in *Poa pratensis*, clones were made of each component in a number of twin pairs. Seven such twin pairs were studied, each individual being represented by 5 clone plants. In this material plant weight and a series of morphological characters were measured. A chemical analysis was also made, thanks to the kind cooperation of Dr. J. LINDBERG.

The chromosome numbers of the twins and the results of the plant weighings are given in Table 10. Due to the high number of chromosomes, the chromosome counts may not be quite accurate. In each twin pair, however, it is quite clear that one member of the twins has about 50 per cent more chromosomes than the other. The pair 3003—7 a and —7 c represents two members of a triplet, in which two plants, a and b, had the normal number while the c-plant had an increased number.

The plants have been cut and weighed in 3 different years and the value for each separate year is the average of the 5 clone plants. Considering first the total production in these 3 years, it is evident that on an average the twins with high chromosome numbers have been less productive than the twins with the lower, normal numbers. This

is true of six of the seven pairs. In one pair, however, the reverse is
true, the average weight of 3645—2 a (2n $= \pm$ 49) being less than half
of the weight of the other twin having 2n $= \pm$ 72. If the total average
values of all twins with high and low chromosome numbers are
calculated the result will be 309 gr. for those with high numbers and
333 gr. for those with normal numbers. The difference is relatively
slight, chiefly due to the unusually high weight of the twin 3645—2 b.
Thus, judging from the above results, *Poa pratensis twins with an in-*

TABLE 10.   *Plant weight in twin clones of Poa pratensis.*

Clone No.	Chromosome number (±)	Average weight in			Total
		1937	1938	1939	
3003—3 a ...	48	82	212	121	415 gr.
b ...	72	80	162	110	352
3003—4 a ...	48	66	171	106	343
b ...	72	66	172	100	338
3003—7 a ...	49	74	148	93	315
c ...	71	50	118	67	235
3009—1 a ...	68	82	144	110	336
b ...	100	25	82	93	200
3839—6 a ...	49	88	170	100	358
b ...	76	46	124	97	267
3839—10 a...	52	76	180	115	371
b...	72	80	162	101	343
3645—2 a ...	49	33	85	78	196
b ...	72	64	208	157	429

*creased chromosome number will in most cases, but not always, be
less productive than the normal twins.*

This conclusion, however, is only based on weight values from
3 years. If the observations had been extended over a longer period
the values would probably have been more favourable for the twins
with high numbers. As already mentioned above, and as is typical of
other polyploids (cf. MÜNTZING, 1936), *the twins with high chromo-
some numbers need more time to reach their full development.* This
is, indeed, evident from a comparison between the weight values of the
different years. Taking first a separate typical twin pair, the weight rel-
ation between the plants 3009—1 a (2n $=$ 68) and 3009—1 b (2n $=$ 100)
was found to be 82 : 25 $=$ 3,28 in the first year, 144 : 82 $=$ 1,76 in
the second year and 110 : 93 $=$ 1,18 in the third year. If this tendency

TABLE 11. *Morphological data from twin clones of Poa pratensis.*

Clone No.	Chromosome number (±)	Height	Culm diameter	Leaf breadth	Leaf length	Thickness of 10 leaves	Panicle length	Spikelet length	1000-grain weight
3003—3 a	48	90,8	1,03	3,70	46,6	2,31	10,25	5,03	0,377
b	72	89,5 —	0,99 —	4,20 +	44,6 —	2,56 +	10,01 —	4,78 —	0,488 +
3003—4 a	48	83,5	0,84	3,60	44,9	2,23	9,45	4,83	0,444
b	72	79,5 —	1,08 +	3,83 +	42,1 —	2,52 +	11,85 +	4,69 —	0,417 —
3003—7 a	49	77,1	0,86	3,79	36,2	2,24	8,96	5,43	0,487
c	71	71,1 —	1,08 +	4,75 +	34,9 —	2,79 +	9,14 +	5,05 —	0,491 +
3009—1 a	68	85,1	0,93	3,70	34,5	2,39	10,15	4,45	0,297
b	100	65,6 —	1,08 +	4,35 +	35,3 +	2,92 +	9,10 —	4,13 —	0,498 +
3839—6 a	49	74,9	0,86	3,60	40,7	2,36	11,67	4,45	0,310
b	76	83,8 +	1,17 +	4,05 +	39,8 —	3,01 +	13,40 +	4,35 —	0,459 +
3839—10 a	52	84,7	0,86	3,80	43,0	2,34	9,35	5,17	0,417
b	72	89,2 +	0,99 +	3,90 +	42,9 —	2,49 +	12,51 +	4,63 —	0,514 +
3645—2 a	49	81,7	0,95	3,85	40,1	2,34	11,30	6,17	0,466
b	72	81,2 —	0,99 +	4,10 +	46,7 +	2,62 +	12,30 +	6,48 +	0,482 +

¹ The + and — signs indicate that in the twins with the higher chromosome numbers the characters have higher or lower values than in the corresponding twins with low chromosome numbers.

continues, the twin with the high chromosome number may in the long run be more productive than the other twin.

Comparing the entire material in the same way, the average weight relation (low : high) was found to be 71,6 : 58,7 = 1,22 in the first year, 158,6 : 146,9 = 1,08 in the second and 102,9 : 103,3 = 1,00 in the third year. Thus, after 3 years the average yield was about the same in the two chromosomal categories.

b. *Morphological data.* — The clone material studied seemed to be suitable also for morphological comparisons between the twins. Therefore the following characters were measured: height, thickness of

TABLE 12. *Chemical data from twin clones of Poa pratensis.*

Field number	Water (per cent)		Crude protein [1] (per cent)		Crude fat [1] (per cent)		Soluble carbohydrates [1] (per cent)		Crude fibre [1] (per cent)		Ashes [1]	
	2 n	3 n	2 n	3 n	2 n	3 n	2 n	3 n	2 n	3 n	2 n	3 n
3003—3 a and b	6,7	6,7	6,7	5,5	3,0	2,2	38,1	38,1	30,1	32,2	7,1	7,0
3003—4 a and b	6,7	6,7	5,7	7,1	2,2	3,2	38,4	37,9	32,0	28,8	6,6	8,0
3003—7 a and c	6,6	6,6	7,3	6,2	2,8	2,9	37,6	38,7	29,4	28,4	7,9	8,9
3009—1 a and b	6,5	6,7	7,3	8,4	2,6	2,0	37,3	37,0	28,2	30,2	9,6	7,4
3839—6 a and b	6,8	6,9	6,8	6,0	2,6	2,2	39,3	38,4	27,6	29,9	8,8	8,6
3839—10 a and b......	6,7	6,6	9,5	5,7	2,6	2,3	35,7	41,7	27,6	24,5	9,7	10,9
3645—2 a and b	6,4	6,6	6,7	7,3	2,4	2,8	39,5	37,2	28,5	28,3	7,9	9,4
Average values:	6,63	6,69	7,00	6,60	2,60	2,51	38,0	38,4	29,1	28,9	8,23	8,60

the culm, length and breadth of the leaves, thickness of 10 leaves put together, panicle length, length of spikelets and, finally, 1000-grain weight. The results of the measurements are summarized in Table 11. With the exception of seed weight, which was only measured once, the values in the table are mean values of measurements undertaken in 1937 and 1938.

Some of the characters measured are clearly correlated with chromosome number, others seem to be more or less independent. The best diagnostic characters were found to be thickness and breadth of the leaves. *In all seven twin pairs the members with high numbers were found to have thicker and broader leaves than the corresponding twins with low numbers.* Also with respect to some of the other

[1] Water content at the analysis = 15 per cent.

characters there are clear differences between the two categories. Thus, high chromosome number is also correlated with thick culm and heavy seeds in six cases of seven. On the other hand, there is a clear negative correlation between chromosome number and length of the spikelets. With respect to the other characters the correlations are less clear. However, the high-number twins seem to be less tall and to have shorter leaves but longer panicles than the sister twins with low chromosome numbers.

c. *Chemical data.* — Thanks to Dr. J. LINDBERG, the same twin material was analysed chemically, the following properties being studied: water content, percentage of crude protein, crude fat, soluble carbohydrates, crude fibre and ashes. The results of this analysis are given in Table 12. The percentage values of the chemical constituents are calculated for a water content of 15 per cent.

The table reveals that *there are no significant differences between high and low chromosome number twins in any of the properties analysed.* In some pairs the number with the higher chromosome number represents the higher percentage values, in other cases the reverse is true. Thus, in this material, in contrast to several other polyploids (cf. MÜNTZING, 1936), the increase in chromosome number does not seem to be accompanied by any obvious chemical alterations.

d. *Pollen fertility.* — Pollen fertility was studied in the seven pairs of clones, the following results being obtained:

Plant number	Chromosome number	Average pollen fertility (per cent good pollen)	Number of samples
3003—3 a	48	80	2
—3 b	72	96	2
—4 a	48	82	3
- 4 b	72	86	1
—7 a	49	83	2
- 7 c	71	97	2
3009—1 a	68	92	2
—1 b	100	93	1
3839—6 a	49	88	2
—6 b	76	96	2
—10 a	52	65	2
—10 b	72	97	2
3645—2 a	49	61	2
—2 b	72	94	2

Though the number of pollen samples taken is low, the result is quite clear, *the twins with high chromosome numbers all having better pollen fertility than the corresponding twins with low numbers.* In one case the difference is incidental rather than significant. The pair 3009—1 a and b has the chromosome numbers 68 and 100, the corresponding percentage values being 92 and 93 respectively. In this pair, however, even the component with lower number has about as high a chromosome number as the high number twins in the other pairs. In this twin pair both members may consequently be regarded as having surpassed the threshold value of chromosome number necessary for a good pollen quality.

e. *Chromosomal variation in twin progenies.* — Some progenies of the twin material studied were raised, primarily, in order to test whether the twins with high chromosome numbers would breed true, or whether they would split up their chromosome numbers. The latter alternative was found to be true. The material in question has not yet been studied morphologically, but the results of chromosome counts in some progenies are available.

In the progeny of twin plant *3003—3 a,* having $2n = \pm 48$, the chromosome numbers of 29 plants were determined, and all of these were found to have numbers ranging from 48 to 51. According to the counts, 6 plants had $2n = 48$, 4 plants $2n = 49$, 16 plants $2n = 50$ and 3 plants $2n = 51$. Considering the difficulties of exact counts, this progeny is probably constant in chromosome number. In such a case the correct chromosome number of the biotype must be $2n = 50$.

The sister twin, *3003—3 b,* having $2n = \pm 72$, gave rise to a progeny in which the chromosomal variation was as follows:

Chromosome numbers: ..	48	49	76	77	80	81	82	83	84	85
Number of plants:	1	1	1	1	6	—	13	6	4	4

Even if the difficulties of exact counts are admitted, it is obvious that chromosomal variation in this progeny is greater than in the offspring of the sister twin with $2n = \pm 48$. The number most frequently represented is as high as 82, and on an average the progeny has a higher chromosome number than the mother plant. The reason of this unexpected increase is not known.

Progenies of two other twin pairs were studied in the same way. In the first of these pairs the chromosome numbers had been determined to be ± 49 and ± 71 respectively. In the offspring of the first plant (*3003—7 a*), 23 plants were found to have numbers ranging

from 47 to 52. Probably, most or all of these plants had in reality the same chromosome number. As in the corresponding member of the previous twin pair and its progeny, 2n = 50 seems to be the typical number also in this case. In 12 of the 23 plants the counts resulted in this number. In addition to the 23 plants having ± 50 chromosomes, there was one single plant with 2n = ± 75 and a twin pair, the members of which were found to have 2n = ± 76 and ± 75 respectively. Thus, a few unreduced ovules had evidently been fertilized.

In the other twin progeny (from the mother plant *3003—7 c,* having 2n = ± 71) the chromosomal variation again seems to be greater, the following numbers being found:

Chromosome numbers:	70	71	72	73	74	75	76	77	78	...	90	...	110	111
Number of plants:	3	–	1	–	5	11	15	6	5	–	1	–	1	2

The plants with 110 and 111 chromosomes are evidently due to the functioning of unreduced ovules. Two of them represent one of the members in each of two twin pairs.

In the third pair of twin progenies variation was again observed in the offspring of the twin with the higher number, the other progeny being almost constant. The mother plants in this case were *3839—10 a* (2n = ± 52) and *3829—10 b* (2n = ± 72). The former individual gave a progeny in which 44 plants were found to have chromosome numbers ranging from 47 to 52. As in the previous cases, 50 was the most frequent number, the counts giving this value in 19 of the plants. In addition to the plants having 2n = ± 50, there was a twin plant with 2n = ± 74.

In the other progeny (from *3839—10 b*) chromosome variation was very marked, the values ranging from 33 to 103. The following numbers were obtained:

Chromosome numbers:	33	...	37	38	...	64	65	66	67	68	69	70	71	72	...	75	76	...	103
Number of plants:	1	-	1	1	—	1	-	1	1	4	—	1	--	9	—	2	3	—	1

In this family the three plants having 33, 37 and 38 chromosomes may be regarded as haploids, having approximately half the chromosome number of the mother plant. Two of these haploids were apparently single plants, the third haploid arose as a twin. In the progeny there is also a »triploid» with 2n = 103, and certainly resulting from the union of an unreduced ovule and a reduced male gamete.

In view of all the data presented above, the following conclusions may be drawn: In contrast to the twin plants representing the normal

chromosome number, the »triploid» twins give rise to progenies in which the chromosomal variation is more or less increased. This demonstrates that the mother plants must be partially or purely sexual. Thus, we have reason to believe that *in Poa pratensis a change from the normal chromosome number to the triploid condition involves a change from a predominant apomictic seed formation to a more or less pronounced degree of sexuality.* More information about this change in the mode of reproduction may be gathered by future studies on the morphology of this material.

VI. CHROMOSOME NUMBER AND CELL SIZE.

As in many other genera and species, comprising types with different chromosome numbers, there is a positive correlation between chromosome number and cell size also in *Poa.* NISSEN (1937) has already reported the occurrence of a positive correlation between chromosome number and size of the stomata in *Poa pratensis.* His measurements are of special interest in this connection, since they were performed on my twin clones of *pratensis*, discussed above in the present paper. As a supplement to his measurements the following data from *pratensis* as well as *alpina* may be briefly described.

1. POLLEN SIZE IN POA PRATENSIS.

In 9 different biotypes of *Poa pratensis,* representing 8 chromosome numbers, ranging from 49 to $\pm$ 87, pollen size was measured. The results given in Table 13 demonstrate a clear positive correlation. The average pollen size of the biotype with $2n = 49$ is approximately 27,7 units, the corresponding average of the biotypes with 64 and 68 chromosomes is 30,7, the total average of the group of biotypes with 70 to 75 chromosomes is 32,0 and the biotypes with 85 and 87 chromosomes, finally, reach an average diameter of 35,3 units.

Pollen measurements were also made in the haploid, sexual *Poa pratensis* type described above (p. 155) and in the corresponding type with $2n = 72$. Two pollen samples of each type were studied, and in each sample 200 grains were measured. In the haploid having $2n = 36$, the average values found were $16,50 \pm 0,08$ in the first and $16,46 \pm 0,08$ in the second sample. In the corresponding type with $2n = 72$, the average values of two samples studied were found to be $17,08 \pm 0,08$ and $17,73 \pm 0,08$ respectively. Thus, the two higher values are represented by the type with the higher chromosome number. However,

TABLE 13. *Pollen diameter in Poa pratensis.*

Year	Field No.	Number of grains measured	Average value	Chromosome number
1932...........................	245	62	27,71	49
1933...........................	»	180	27,82	»
»	»	150	27,54	»
»	149	»	28,94	⊢ 64
»	124	»	32,48	± 68
»	124 L	180	31,94	± 70
»	127	»	31,73	72
1932...........................	»	150	32,59	»
»	229	»	31,99	± 75
1933...........................	»	»	32,24	»
1932...........................	126	»	31,39	»
»	150	»	34,83	± 85
1933...........................	158	»	35.73	+ 87

the difference in pollen size between the two types compared is surprisingly small.

2. SIZE OF POLLEN AND STOMATA IN POA ALPINA.

Pollen size was determined in 23 different plants of *Poa alpina*, the chromosome numbers of these plants ranging from 22 to 43. The plants were chosen only with regard to their chromosome number, and thus they belong to sexual as well as apomictic strains or were hybrids of different kinds. From each plant 50 good pollen grains were measured. The following mean values were obtained (the corresponding chromosome number in brackets): 26,2 (22), 26,4 (22), 24,4 (23), 25,1 (24), 25,5 (24), 25,9 (25), 26,1 (25), 28,2 (26), 25,3 (27), 26,2 (28), 28,6 (29), 26,9 (31), 27,9 (31), 27,0 (32), 26,7 (33), 27,2 (33), 26,4 (36), 34,0 (37), 28,8 (38). 29,3 (39), 31,9 (41), 30,5 (43).

The positive correlation between pollen size and chromosome number is not strong but, nevertheless, clear. If the values are combined in classes comprising several chromosome numbers, the following values will be obtained:

Chromosome classes: 22 — 26 -- 30 - - 34 -- 38 — 42 — 46
Average pollen diameter: 25,6   27,1   27,1   30,2   30,0   30,5

According to the above values, the diameter of the pollen grains is about 20 per cent greater in the 42—46 class than in the 22—26 class.

This means that the relation between the volumes of the two classes compared will be $2837 : 1677 = 1{,}7 : 1$. The relation between the average chromosome numbers of the same classes is $1{,}8 : 1$.

In *Poa alpina* length of the stomata was also measured in 26 plants, representing 23 different chromosome numbers, ranging from 22 to 74. Part of these plants were also used for the pollen measurements. From each plant 50 stomata were measured. The following average values were obtained (the corresponding chromosome numbers in brackets):

$12{,}28$ (22), $14{,}00$ (23), $14{,}44$ (24), $11{,}92$ (24), $14{,}36$ (24), $12{,}78$ (25), $17{,}71$ (26), $14{,}90$ (27), $15{,}28$ (28), $14{,}98$ (29), $15{,}70$ (30), $15{,}72$ (31), $15{,}26$ (32), $17{,}30$ (33), $17{,}06$ (34), $13{,}91$ (35), $15{,}44$ (36), $15{,}46$ (37), $15{,}18$ (38), $16{,}52$ (39), $16{,}12$ (40), $19{,}00$ (41), $19{,}08$ (42), $17{,}16$ (43), $20{,}90$ (52), $19{,}30$ (74).

If these values are arranged in a series according to increasing chromosome numbers, using the same class width as for the pollen, the result will be as follows:

Chromosome classes   : 22 — 26 — 30 — 34 — 38 — 42 — 46 — 50 — 54 ... 74
Average stomata length: 13,0 15,7 16,0 15,5 16,7 18,1   —   20,9   —   19,3

In spite of the incidentally low value in the class 34—38 the positive correlation between chromosome number and stomata length is quite clear.

VII. DISCUSSION.

1. *The genotypical basis of apomixis.* — The main problem in the material studied is the genotypical basis of apomixis and the relationship between sexual and apomictic strains. The first fact to be considered in connection with this problem is the occurrence of gradations between sexuality and apomixis. In *Poa alpina* the Fürstenalp strain was found to be highly variable in morphology as well as in fertility and chromosome number and may therefore be regarded as purely sexual. On the other hand, several of the Scandinavian strains have so far been completely agamospermic, not a single aberrant plant being observed in the progenies. Other strains, however, occupy an intermediate position, a certain proportion of aberrants being formed. In the strain from St. Gothard sexuality is rather pronounced, the minimum percentage of aberrants being $25{,}93 \pm 8{,}43$. The strain from Mösseberg is more stable, the percentage of aberrants being as low as $4{,}35 \pm 3{,}01$. Further examples of this kind were met with in the new Swiss collection of *Poa alpina* strains discussed above (p. 138—145). Different

degrees of apomictic seed formation have also been observed in *Poa pratensis,* judging from differences in the frequency of morphological aberrants (ÅKERBERG, 1939).

Already the occurrence of such gradations in the degree of sexuality is a strong indication that apomixis is not conditioned by a single factor. It might be assumed, however, that plants and strains being heterozygous for a single apomixis factor (Aa) might be partially sexual in contrast to the homozygous combinations, aa and AA, which would represent pure sexuality and absolute apomixis. Against this interpretation may be said, firstly, that most probably there is more than one gradation between absolute sexuality and absolute apomixis. Secondly, the cross between sexual and apomictic *Poa alpina* gave F_1 hybrids, which were purely sexual and not intermediate. Finally, the absence of apomictic F_3 families in the same cross may be regarded as definite evidence that in the material studied apomictic seed formation is conditioned by more than one factor.

A curious fact to be considered in this connection is that a change from the normal chromosome number to »haploidy» or »triploidy» seems to involve a change from predominant apomixis to a more or less pronounced sexuality. The haploid *pratensis* type with 36 chromosomes was clearly sexual, in contrast to the original type having the double chromosome number. Also the twin plants, having approximately a 50 per cent higher chromosome number than usual, behaved in a similar way. At least some of these twin plants are decidedly more sexual than the corresponding sister twins, representing the normal chromosome number of the strain. ÅKERBERG (1939) has already reported similar results, the progenies of aberrant *Poa pratensis* plants in his experiments always being variable in morphology as well as chromosome number.

If apomixis is conditioned by multiple factors, the sexuality of a haploid type is rather natural, since such a haploid will only contain part of the factors necessary for an apomictic seed formation. The sexuality of triploids is more difficult to understand but leads to the idea that apomixis in *Poa* is due to a rather delicate genetic balance. This balance may be upset in various ways, by crosses with other types or merely by a quantitative change in chromosome number either in a plus or minus direction.

From the experience gathered in our material there is reason to believe that apomixis is an advantageous property, gradually evolved by natural selection. When grown together in the field the apomictic

strains differ favourably and strikingly from sexual progenies by their uniformly good vigour and good seed production. Only a small proportion of the plants in the sexual strains are comparable to the apomicts in these respects. Further, if aberrants with deviating chromosome numbers are produced in predominantly apomictic but partially sexual strains, these aberrants have often been observed to be less vigorous than the typical plants.

On the other hand, the sexual strains may be supposed to represent a valuable source of new types, some of which have good vigour and other advantageous properties. The preservation of such valuable individuals by apomixis may be easily imagined, since in *Poa pratensis* and *alpina* the tendency to apomictic seed formation is evidently widespread. Even in types considered to be typically sexual a certain proportion of unreduced embryo-sacs may be formed. Thus, in the cross between the sexual plants with $2n = 24$ and the apomictic strain with $2n = 38$, 6 F_1 plants of 202 were found to be the result of unreduced ovules fertilized by reduced male gametes (p. 148). — In the sexual haploid of *Poa pratensis* ($2n = 36$) there is a similar proportion of functional unreduced ovules, three »triploids» being observed among a total of 44 plants. Also in many other sexual or partially sexual *alpina* and *pratensis* plants there is a marked tendency to formation of unreduced gametes, chiefly on the female side but also in the pollen.

If by mutation a vigorous plant belonging to a sexual strain becomes capable of developing its low proportion of unreduced ovules without fertilization, it is conceivable that by secondary changes the proportion of apomictic offspring may be gradually increased. This is probable, especially because the offspring formed in a sexual way will, on an average, be less successful than those possessing all the advantageous properties of the original mother. — The nature of the mutation, responsible for the capability of development without fertilization, is of course a matter of speculation. It may or may not be a true gene mutation. Perhaps apomixis is brought about by quite special constellations of genes and chromosomes. Some of these cause the egg cells to develop without fertilization, others increase the number of unreduced embryo-sacs, the resultant of these factors being the proportion of apomictic offspring.

The apomicts in *Poa alpina* and *pratensis* are peculiar by their aneuploid chromosome numbers, but otherwise the apomictic phenomena in these species are similar in many respects to the con-

ditions in the genus *Rubus*. As demonstrated by LIDFORSS (1905, 1907 a and b, 1914), the *Rubus* species are generally predominantly pseudo-gamous but partially sexual. The hybrids formed, however, seem to be purely sexual and give a very strong segregation in the next generation. On the basis of these results and experiments of his own, GUSTAFSSON (1930) concludes that in *Rubus* apomixis is recessive to sexuality. In *Poa* apomixis was found to be recessive in hybrids between typically sexual and apomictic plants, but sexuality was also induced by other genotypical changes, viz. haploidy and triploidy. In *Rubus*, also, a new genotypical constitution, involving a complex change, always seems to be accompanied by loss of apomictic propagation. The *Rubus* hybrids produced were all sexual, even if both parents were pre-dominantly apomictic. — On the other hand, LIDFORSS (l. c.) observed mutations in many *Rubus* species, leading to new morphological types but not to loss of apomictic propagation. This seems to indicate that apomixis in *Rubus* as well as in *Poa* is conditioned by the cooperation of several or many factors. This cooperation may be upset by crossing or by quantitative chromosome changes but is not disturbed by minor changes such as gene mutations.

In his publications LIDFORSS briefly mentions the important fact that the offspring of hybrids between partially pseudogamous parents show a tendency in later generations to become stabilized again. This probably implies that vigorous segregation products, containing gene constellations for different degrees of apomixis, are favoured at the expense of the purely sexual products. In *Poa alpina* such a stabiliz-ation has not yet been observed, all the F_3 progenies in the cross sexual $\times$ apomictic so far studied showing a marked variation. A return back to apomixis may perhaps occur in some families in later generations.

Crosses between plants representing different degrees of sexuality and apomixis have also been undertaken in *Hieracium* by MENDEL (1869) and OSTENFELD (cf. OSTENFELD, 1910). As is well-known, MENDEL found that his *Hieracium* material behaved in quite the opposite way to the *Pisum* hybrids. The F_1 generation was polymorphic, but each F_1 plant, from which offspring could be obtained, was true-breeding and gave a uniform progeny of maternal type. In OSTENFELD's material the same result was obtained in most cases, but in a few crosses all or part of the F_1 plants were sexual or partially sexual. Thus, in *Hieracium* (subgenus *Pilosella*) apomictic seed formation generally seems to be dominant over sexuality. Recent results in

Potentilla seem to go in the same direction (A. and G. MÜNTZING, un-published data). By crosses between two biotypes of *Potentilla Taber-næmontani* (T—B × T—A; cf. MÜNTZING, 1931) a few hybrids were obtained, the chromosome numbers of the mother, father and F_1 being 84, 42 and 63 respectively. Judging from morphological data and preliminary chromosome counts, the hybrids are true-breeding as in *Hieracium*.

Thus, in *Hieracium* and *Potentilla* crosses between apomictic and sexual or partially sexual strains have given other results than in *Rubus* and *Poa*. In the latter category apomixis is recessive to sexuality, in *Hieracium* and *Potentilla* apparently dominant. Probably, however, this difference is only gradual. In the *Hieracium* crosses sterile or self-sterile hybrids were frequently obtained besides F_1 plants giving apomictic progeny (OSTENFELD, l. c.). These sterile plants may in part represent sexual F_1 plants, and in such a case the dominance of apomixis is not complete. On the other hand, the F_1 plants in the cross sexual × apomictic *Poa alpina* had a tendency to produce haploids, indicating an apomictic tendency to development without fertilization (cf. below p. 181).

2. *Groups of Poa alpina apomicts with a different geographical distribution.* — Though the apomictic strains of *Poa alpina* are charact-erized by peculiar aneuploid chromosome numbers, it is a striking fact that a particular chromosome number is characteristic of groups of biotypes occupying a special geographic region. Thus, the most fre-quent number among the Swiss apomicts is *37*, this number being characteristic of 7 strains from 5 different localities. So far this num-ber has not been met with in the Scandinavian material, which is characterized by the numbers *38* and *33*. The former number was found in biotypes from the provinces of Jämtland and Norrbotten (Sweden) and in a type from Troms (Norway). The number 33 was represented by one biotype from Norrbotten and by the biotypes from Gotland and Mösseberg (South Sweden). The number 33 was also met with in one of the Swiss strains and in a viviparous biotype from arctic Norway (FLOVIK, 1938). The same author found two viviparous *alpina* biotypes from Spitsbergen to have 2n = 44 and 2n = 42 + 4 ff. Finally, the biotype from Öland (Sweden) represents a special number, 2n = 35.

It is evident that the chromosome numbers of the apomictic strains have a special significance, and that they are not merely chance num-bers in.a material showing the same great and continuous variation as the sexual strains with oscillatory chromosome number. It is obvious

that the Swiss strains with $2n = 37$ are more related to one another than to strains having different chromosome numbers. It seems highly probable, also, that among the Scandinavian biotypes the members of the group with $2n = 38$ are more related to each other than to the members of the group with $2n = 33$. The latter group is especially interesting from a plant-geographical point of view, since it comprises northern types (from Pajala and arctic Norway) as well as two south Swedish biotypes (Mösseberg and Gotland).

TURESSON (1927) has discussed the relationship between south and north Swedish *Poa alpina* and especially the supposed relic nature of the former. He comes to the conclusion that the Scandinavian population of *Poa alpina* is split into three different ecotypes, one alpine, one subalpine and one lowland ecotype. Further, the conclusion is drawn that the lowland occurrences of *Poa alpina* do not represent glacial relics, descending from the first immigrants of the species but are later types, having immigrated into Sweden when the climate had become more favourable. — Now the situation in *Poa alpina* has become rather changed, since it has been found that the species in Scandinavia is represented by apomictic biotypes characterized by quite special chromosome numbers. The biotypes from northern Scandinavia, studied by me, seem to correspond morphologically to the subalpine ecotype described by TURESSON. My material, however, was found to comprise two different chromosome numbers, 38 and 33, the latter number also being found in two of the three south Swedish biotypes studied. This strongly indicates that there are biotypes in the northern part of Scandinavia, which as regards their ultimate origin are of the same kind as the strains from Mösseberg and Gotland. — Though the evidence now available is too meagre for much speculation, I would imagine that the first immigrants of *Poa alpina* into Sweden belonged to the 38-chromosome group, and that these were followed by biotypes having $2n = 33$. Some of them were capable of going far to the north, others settled down in the southern parts of this country.

Morphologically all the *alpina* strains so far studied by me were found to be different, the most deviating type being the strain from Gotland (Fig. 15). No clear average difference in morphology between the groups with 38 and 33 chromosomes could be observed. Nevertheless, I should think that the members within each group are of a common origin, and that the morphological and ecological differentiation has occurred after these peculiar chromosome numbers

were established. Theoretically it is possible that the Scandinavian apomict groups with 2n = 38 and 33 have been differentiated from one original immigrant biotype with 38 and another biotype with 33 chromosomes. At any rate, it is very probable that the Mösseberg and Gotland strains have relatives in North Scandinavia. Though improbable, the possibility is not quite excluded, however, that the south Swedish *alpina* types are really of a more recent origin. A single Swiss agamospermic strain was, indeed, found to have 33 as its typical chromosome number, though most of the other Swiss strains had 2n = 37.

The complexity of the problem is also illustrated by the fact that the Öland strain of *Poa alpina*, studied by me, had a quite special chromosome number, 2n = 35, not met with in any other *Poa alpina* type. Judging from TURESSON's descriptions, there must also exist several different *alpina* biotypes in Öland, his type being much more vigorous than mine. — Evidently, much more work is needed before a deeper insight into the problems outlined here can be gained. The most interesting problem at present seems to be whether the *Poa alpina* types in Scandinavia and other regions are really divided into definite groups, characterized by special chromosome numbers and special geographical distribution areas.

3. *The formation of new biotypes and the origin of vivipary.* — With respect to the Scandinavian *alpina* biotypes it was assumed that a differentiation may occur by mutation, which does not involve a change in chromosome number. More evidence in favour of this interpretation was obtained from a study of the agamospermic and viviparous biotypes from Switzerland. Among the group of strains having 2n = 37 as their characteristic chromosome number, all biotypes were, with one exception, morphologically different. The exception is represented by a strain from Arosa, which was indistinguishable from a strain collected at Rigi. A third strain, from Oberalp, was also very similar, but differed in some minor respects. These observations certainly indicate that a morphological differentiation may occur due to mutations, not affecting the chromosome number.

It should be remembered, of course, that in the Swiss material, including some strains with 2n = 37, different degrees of partial sexuality were met with. This might, indeed, be expected to contribute to a differentiation of the species in question. In most cases such a differentiation should lead to the establishment of new chromosome numbers, and we have reason to believe that most of the products formed in this way would not stand the test of natural selection. On the

other hand mutations, not affecting the chromosomal balance, which guarantees a good vigour and a predominant apomictic seed formation, would have a much better chance to lead to new biotypes with a good survival value.

Chromosome counts in two viviparous clones from Switzerland demonstrated with a high degree of probability that differentiation by mutation may not only lead to more or less conspicuous morphological differences but also to a transition from agamospermy to vivipary. — The chromosome numbers of the viviparous clones were found to be $2n = 26$ and $2n = 33$ respectively. These clones were both collected at Arosa. From the same locality five agamospermic or partially sexual strains were also gathered, three of them having $2n = 37$, one strain $2n = 33$ and one strain $2n = 26$ as their typical chromosome numbers. The numbers 26 and 33 were not met with in any of the remaining biotypes from Switzerland. This being the case, it may safely be assumed that the viviparous clones with $2n = 26$ and $2n = 33$ must be rather closely related to the seed producing strains, having the same unique chromosome numbers and being collected at the same locality. Since the chromosome numbers are unchanged, it also seems probable that the viviparous clones have arisen by mutation from the corresponding seed-producing biotypes. — In this connection it should also be observed that the viviparous *Poa alpina* type from arctic Norway (FLOVIK, 1938) had the same chromosome number ($2n = 33$) as several of the agamospermic Scandinavian strains.

4. *Is apomixis in Poa caused by hybridization?* — In his book on apogamy, ERNST (1918) considers the possibility that the viviparous forms of *Poa alpina* might be the result of species hybridization, and FLOVIK (1938) believes that they are allopolyploids. As demonstrated above, however, the evidence now available strongly suggests that the viviparous types are differentiation products, arisen by mutations from non-viviparous strains. This mode of origin seems to be somewhat continuous. ERNST (l. c.) describes the occurrence of semiviviparous types in Switzerland (*Poa alpina* f. *intermedia*). — On account of various observations, NANNFELDT (1937 b) also concludes that vivipary is not necessarily the result of hybridization. — Thus, though the viviparous forms of *Poa alpina*, as contrasted with the agamospermic ones, cannot be regarded as species hybrids, this does not exclude the possibility that in other genera viviparous types may arise in such a way. FLOVIK (1938) has presented evidence, based on chromosome morphological studies, that two different viviparous *Festuca ovina*

types from Spitsbergen and arctic Norway should have arisen through crosses between certain varieties of *F. rubra* and *F. ovina*.

In *Crepis* and *Antennaria*, on the contrary, BABCOCK and STEBBINS (1938) report results which are considered to indicate strongly that the onset of total apomixis in these genera is a gradual process, facultative apomixis often preceding the obligate type. The same authors also find that in *Crepis* hybridization is not the cause of apomixis, many auto-polyploids being just as completely apomictic as are the allopolyploids.

Though vivipary in *Poa alpina* cannot be considered to be the immediate result of hybridization, the possibility must not be overlooked that the whole species complex, *Poa alpina*, might be of a hybrid origin. In such a case agamospermy, and secondarily vivipary, might ultimately be based on the genotypical changes brought about by hybridization. In *Rubus* the groups of apomictic species are considered to be the result of species hybridization (GUSTAFSSON, 1930). — The evidence available in *Poa alpina*, however, is not in favour of such an explanation. The meiotic observations demonstrate a rather marked degree of auto-polyploidy rather than allopolyploidy. In the first place this is evident from a relatively high frequency of trivalents and larger associations in most of the plants studied. Secondly, the very good pollen fertility in the aneuploids and the absence of a correlation between chromosome number, vigour and fertility point in the same direction. In these respects the *Poa alpina* material is similar to pentaploid *Dactylis* plants (MÜNTZING, 1938 a). On account of their autopolyploidy such plants have quite good pollen, in contrast to their triploid mothers. In the offspring there is no correlation between chromosome number and pollen fertility and only a weak correlation between chromosome number and vigour.

In *Poa pratensis* higher chromosome numbers than in *Poa alpina* must be reached in order to secure a quite good pollen fertility. As a rule, *pratensis* plants having about 50 chromosomes seem to be partially sterile, but as soon as the chromosome numbers exceed 70, approxim-ately, the pollen is perfectly normal. This was especially evident in the diplo-triploid *pratensis* twins and may also be observed when differ-ent *pratensis* strains are compared (MÜNTZING, 1932; ÅKERBERG, 1936 a). This indicates that in *Poa pratensis* there is more differentiation be-tween the genomes than in *Poa alpina*, or in other words that auto-polyploidy is more pronounced in *Poa alpina* than in *pratensis*. In *Poa serotina* (= *palustris*), finally, KIELLANDER (1935, 1937) has shown that a tetraploid apomictic strain, studied by him, is probably auto-

tetraploid, 3—4 quadrivalents generally being present at first meta-phase.

5. *The aneuploid chromosome variation.* — The variation in chromosome number within the species *Poa alpina* and *Poa pratensis* shows a good deal of resemblance to the aneuploid chromosomal variation within the genus *Carex*. In a recent paper, HEILBORN (1939) agrees with the view advanced by MEURMAN and myself (cf. MÜNTZING, 1936, p. 361) that part of the aneuploidy in *Cyperaceæ* is probably a result of meiotic instability in autopolyploids, part of the aneuploidy in the genus thus being the result of a breakdown of original autopolyploid forms. In *Carex* the aneuploid variation in chromosome number is correlated with species differentiation. In *Poa*, on the contrary, there is at least in part a true intraspecific variation in chromosome number. Certainly it is not possible, for instance, to separate the Scandinavian *alpina* apomicts into three different species, characterized by the chromosome numbers 33, 35 and 38. — The Swiss strains of *Poa alpina* studied have not been subjected to any detailed morphological comparison with the Scandinavian representatives of the species. However, there does not seem to be any obvious morphological characters distinguishing the two groups. At any rate they must be rather closely related, the hybrids between a Swiss and a Scandinavian strain having quite good fertility.

As already demonstrated in my previous paper on *Poa* (MÜNTZING, 1932), the apomictic strains are true aneuploid forms, and it is not possible to explain the deviations from multiples of seven merely by the assumption of fragmentation and other structural changes. Further evidence pointing in the same direction has been obtained from the meiotic studies now reported. As is evident from their size, the un-paired chromosomes, frequently occurring at meiosis in *Poa alpina*, are obviously true univalents and not fragments. — In spite of the absence of correlations between chromosome number, vigour and fertility the *Poa alpina* chromosomes are probably not inert, progenies with variable chromosome numbers always showing a marked pro-portion of plants with poor vigour and fertility.

Further evidence that the intraspecific differences in chromosome number really represent different chromatin quantities is represented by the observed positive correlations between chromosome number and cell volume. In *Poa alpina* as well as in *pratensis* the size of the pollen grains was found to be proportional to the chromosome number, and in *Poa alpina* the same thing was found to be true of stomata size.

Though, consequently, there must be a true aneuploid variation in chromosome number in the two species, this does not exclude the possibility that this aneuploidy may have been further changed by structural differences of various kinds. Indeed, in *Poa alpina*, as well as in the haploid *pratensis*, dicentric chromatids and accompanying small fragments were observed. Thus, the plants in question must have been heterozygous for inversions or duplications.

The observed absence of a correlation between chromosome number, vigour and fertility in *Poa alpina* explains the chromosomal polymorphism of the species, occurring in nature. Not only the apomictic strains are successful with aneuploid chromosome numbers, but also in sexual material of the species the chromosome number may evidently remain oscillating for a long time, since no selective forces tend to decrease this chromosomal variation. — When comparing the F_1 and F_2 plants in the cross sexual $\times$ apomictic *alpina*, the chromosomal variation was found to be just as intensive in F_2 as in F_1, and there was not even an average decrease in chromosome number. Such a decrease, however, was observed in progenies of the sexual strain from Fürstenalp, and theoretically it should be expected in all offspring from mother plants having an irregular meiosis.

Though no correlation between chromosome number and percentage of apparently good pollen grains could be established, a certain degree of selective gametic elimination may nevertheless be suspected to occur. This selection will probably favour gametes with numbers higher than the average. In this way the inevitable chromosomal elimination may be counterbalanced and in some cases even lead to progenies in which most of the plants have a higher chromosome number than the mother. A marked example of this kind is represented by one of the progenies of the »triploid» *pratensis* twins.

Even if the chromosome number has a decreasing tendency, as is undoubtedly the case in some sexual *alpina* strains, the chromosomal polymorphism will be upheld, due to the occasional functioning of unreduced gametes. The union of unreduced ovules and reduced male gametes was, indeed, observed to be quite a common feature in sexual or partially sexual *alpina* progenies. In several cases even tetraploids were obtained, male as well as female gametes being unreduced. Especially if the functioning of unreduced ovules is repeated in consecutive generations, as was observed to be the case in one of the Swiss *alpina* strains, a very considerable change in chromosome number may result.

From the »triploids» and »tetraploids» arisen by the functioning of unreduced gametes large numbers of new forms with varying chromosome numbers will certainly arise, and no permanent chromosomal equilibrium will be attained.

6. *The spontaneous formation of haploids.* — Chromosomal variation in *Poa alpina* and *pratensis* is not only characterized by »jumps» in the plus direction, eventually followed by a slow process of decrease in chromosome number. The opposite process may also occur, viz. the sudden formation of approximate haploids, having about half as many chromosomes as their mother plants. In the first place a regular formation of haploids was observed in the offspring of certain hybrids between sexual and apomictic *alpina*. These hybrids had a relatively high chromosome number $(2n = 41)$ and were the result of a union between unreduced ovules and reduced male gametes. In the progeny most closely studied the percentage of haploids formed was as high as 14,8 (Table 9). In the material in question this formation of a rather high proportion of haploids may be explained, formally, in the following way.

From the sexual parent the F_1 plant has inherited a tendency to produce reduced gametes and from the apomictic parent a tendency to development without fertilization. These tendencies combined may be responsible for the parthenogenetic development of reduced ovules. — It is rather probable that the F_1 plants with lower, ordinary chromosome numbers have the same tendency, but in these cases the chromosome numbers of the haploids will be too low, the resulting individuals not being viable. — This idea is supported by the fact that in the sexual *alpina* strain from Fürstenalp selection for low chromosome numbers did not result in plants with lower numbers than 21. — In *Dactylis* the effect of unbalanced chromosome numbers has also been observed to be much more serious, when the absolute chromosome number is low than when it is high (MÜNTZING, 1938 a).

Spontaneous production of a haploid was also observed in another progeny of *Poa alpina* (p. 144). This progeny belonged to a Swiss strain, which was evidently partially sexual. It comprised plants with several different chromosome numbers, the most typical ones being 26 and 52. Also in this case a combination of two tendencies may be assumed, one tendency to reduction, another to parthenogenesis. Usually, reduction is combined with fertilization and non-reduction with parthenogenesis, but evidently these variables may sometimes be recombined. In such cases the result may be either triploids or haploids.

It should be remembered that haploids were not only produced in *Poa alpina* but also in *Poa pratensis*. In this species the haploids generally seem to arise as twin plants. — Thus, in both the species studied chromosomal variation may evidently go in two directions, from low to high numbers as well as in the opposite direction. This process of doubling and halving in combination with the frequent occurrence of different degrees of apomixis explains the extreme variation in chromosome number, which is typical of both species.

7. *Embryology.* — A more complete picture of the situation in *Poa* will be obtained when more thorough embryological investigations have been performed. One fact, which is clear already now, is that the apomictic mechanism in *Poa alpina* and *pratensis* is different, at least in the strains studied so far. In *Poa pratensis* there is evidently apospory (ÅKERBERG, 1939), in *Poa alpina*, on the contrary, diploid parthenogenesis (Figs. 18—19). Thus, the *Poa alpina* apomicts are characterized by the same embryological mechanism as *Poa serotina* (KIELLANDER, 1935). In the latter species haplo-parthenogenesis was also observed, and even fertilization may evidently occur (KIELLANDER, l. c.). — It is rather interesting that different *Poa* species have solved the acquisition of an agamospermic mechanism in various ways, which are certainly controlled by different gene constellations. This is probably the result of a gradual process. Two different constellations of factors have finally been established, which satisfy the need of an apomictic seed formation in two different ways.

8. *Secondary polyploidy.* — A rather striking cytological fact, observed in the present material, is the establishment of a balanced secondary polyploidy in *Poa alpina* as well as in *Poa pratensis*. In the former species selection for low chromosome numbers in the sexual Fürstenalp strain led to the production of a stable, but still sexual strain having $2n = 22$. At meiosis 11 bivalents were regularly formed and, on an average, 84 per cent of the plants in the progenies had $2n = 22$ like their mother plants. Since the original basic number in *Poa* is certainly 7, the transition to a new basic number of 11 is really remarkable. — This case deserves further study, especially as regards the possible occurrence of secondary association. So far, however, such an association has not been observed. It would also be interesting to study this stable strain from a chromosome morphological point of view and make comparisons with diploid *Poa* species having $2n = 14$ (e. g. *Poa trivialis*).

Another case of secondary polyploidy was met with in the sexual,

(poly-)haploid *Poa pratensis* having 2n = 36. To my surprise meiosis in this type was highly regular, 18 bivalents being the typical I—M configuration. The regularity of meiosis was also proved by the predominant chromosomal constancy in the offspring after isolation. 72 per cent of the plants had 2n = 36 like their mother, and of the remaining plants 17 per cent had numbers close to 36 (p. 158). After open pollination, on the other hand, chromosomal variation in the offspring was very strong, due to crosses with other *pratensis* biotypes having various high chromosome numbers.

When speaking of the *alpina* and *pratensis* types under consideration as secondary polyploids, it should be kept in mind, however, that their chances to survive under natural conditions are somewhat dubious. The *pratensis* strain, especially, is handicapped by its rather pronounced degree of sterility, though vigour is quite good. The *Poa alpina* type with 2n = 22 has a much better fertility and also a good vigour. Since this strain was produced by selection, however, and not taken directly from nature, nothing definite can be said about its survival value in competition with other strains. — Nevertheless, the striking changes of the apparent basic chromosome number are of interest and strongly support the conclusions drawn by previous workers (cf. DARLINGTON, 1937, pp. 239—243) that genera with high basic numbers, such as *Pyrus, Gossypium, Salix* and *Populus,* are derived from ancestors having lower basic chromosome numbers.

9. *Haplontic and diplontic sterility.* — When considering sterility in the sexual, polyhaploid *pratensis* type just discussed, it should be observed that this sterility is certainly diplontic (or genic, according to DOBZHANSKY's terminology; DOBZHANSKY, 1937). Meiosis is regular, and the pollen quality rather good, but seed production very low. — Diplontic sterility may also occur in *Poa alpina.* In the Fürstenalp strain with oscillating chromosome number part of the plants were found to be male sterile, the anthers not dehiscing. This male sterility was positively correlated with the degree of tillering, vigorous plants with many shoots generally having dehiscing anthers. Thus, sterility of this kind is controlled by the somatic condition of the mother plant. However, haplontic sterility is certainly also at work in *Poa.* This is indicated by the fact that in *Poa pratensis* plants with high chromosome numbers have a better pollen fertility than plants with low numbers. In the former all the pollen grains obtain a sufficient amount of factors necessary for their viability, in the latter part of the pollen grains are killed due to their own unbalanced constitution.

10. *Conclusion.* — The rather complicated cyto-genetical phenomena met with in *Poa alpina* and *pratensis* are evidently not limited
to these species alone but are also characteristic of a rather large group
of other *Poa* species. The occurrence of diploparthenogenesis in *Poa
palustris* (= *serotina*) has already been referred to above. The same
species was also found to comprise strains with different chromosome
numbers (triploid and tetraploid) and strains showing different degrees
of apomictic seed formation (KIELLANDER, 1935, 1937). The definite
or probable occurrence of apomictic seed formation in *Poa* has been
extended by FLOVIK (1938) to the species *alpigena, glauca* and *arctica.*
Poa alpigena was found to comprise strains with different chromosome
numbers, and meiosis was found to be irregular in representatives of
all three species. Thus, the results described in this paper are not
unique within the genus but may be obtained in other *Poa* species as
well. The data from *Poa alpina* and *pratensis* now published may be
regarded as one of the contributions to an analysis of this interesting
genus, which is now being attacked by various workers by means of
cyto-genetical, embryological, morphological and plant-geographical
methods. — The main problem in the genus seems to be the origin
and genetic basis of apomixis. By combined efforts it should be
possible to elucidate this problem rather thoroughly, the material available being very well suited for such studies. In the present paper the
discussion of the apomictic phenomena has chiefly been confined to
the genus *Poa* itself. When more data from the *Poa* species have been
gathered the discussion may be extended to the apomictic phenomena
in general.

VIII. SUMMARY.

1. Of 8 different apomictic strains of *Poa alpina* 4 were constant
in morphology and chromosome number. In the other four strains
different frequencies of aberrants were observed. In most strains the
frequency of aberrants was not increased by mixed pollinations.

2. The *Poa alpina* apomicts are characterized by various aneuploid
chromosome numbers. Biotypes from the same geographical region
tend to have the same number. Thus, most of the Swiss apomicts had
2n = 37, the Scandinavian strains representing the numbers 33, 38 and
35. The characteristic chromosome numbers are valuable for plant-
geographical studies.

3. Meiosis in some *Poa alpina* apomicts was found to be irregular.
The frequent occurrence of multivalents, combined with the fact that

the strains studied have a very good pollen fertility, indicates a rather pronounced degree of autopolyploidy. There was also evidence of a structural differentiation of the chromosomes.

4. Diploid parthenogenesis according to the *Antennaria*-scheme was observed in one of the Swedish apomicts of *Poa alpina.*

5. In a sexual *alpina* strain with oscillatory chromosome number individual progenies showed a slight average decrease in chromosome number, the aberrants with lower numbers than the mother being more numerous than those with higher numbers. In this material there was no correlation between chromosome number, vigour and fertility but a positive correlation between degree of tillering and male fertility. Meiosis in this sexual strain is of the same type as in the p. m. c. of the apomictic biotypes.

6. In the sexual *alpina* strain selection for high and low chromosome numbers was undertaken. Selection for high numbers gave offspring with chromosome numbers ranging from 25 to 64. In relation to the chromosome number of the mother plants the majority of these plants were minus variates. Due to the functioning of unreduced gametes the degree of chromosome variation among the plus variates was stronger than among the minus variates. Also in this material no correlation between chromosome number and vigour could be established.

7. Selection for low chromosome numbers gave rise to a stable, but still sexual strain with 22 chromosomes. Meiosis in this strain is regular, 11_{II} being present at I—M.

8. Another case of secondary balance was met with in a sexual polyhaploid of *Poa pratensis* having $2n = 36$. At meiosis in this type 18_{II} were present at I—M and most of the offspring after isolation had $2n = 36$ like the mother.

9. In a sexual *alpina* plant, giving tetraploid offspring, the pollen size curve was found to be bimodal in some samples, unimodal in another one. Unreduced pollen grains were evidently formed only under certain environmental conditions.

10. A collection of *Poa alpina* strains from Switzerland were studied. Predominantly, this material was found to be apomictic, but some strains were sexual to varying degrees. The most frequent deviations in chromosome number were due to the functioning of unreduced gametes. Also the opposite process was observed, viz. the formation of haploids from types with high chromosome numbers.

11. On account of their chromosome numbers two viviparous

strains studied must be closely related to agamospermic strains from the same locality. In material with the same peculiar and aneuploid chromosome number a differentiation by mutation must be assumed, leading to morphological diversity or even to a transition from agamospermy to vivipary. It is not excluded, however, that the whole species complexes *Poa alpina* and, especially, *Poa pratensis* are of a hybrid origin.

12. Crosses between a sexual and an apomictic *Poa alpina* strain were undertaken. The chromosome numbers of the parents were 24 and 38. In F_1 the chromosome numbers ranged from 25 to 43, one maximum corresponding to reduced female + reduced male gametes, another maximum to unreduced female + reduced male gametes.

13. The F_1 plants had good fertility and were found to be quite sexual. Of 17 F_2 progenies raised, all showed variation in morphology as well as in chromosome number. A regular formation of haploids (15 per cent) was observed in the offspring of some F_1 plants with high chromosome numbers. This must be due to a combination of the parental tendencies to chromosome reduction and parthenogenetic development.

14. All F_2 plants tested were found to be sexual. Therefore, apomixis cannot be due to a single gene but rather to special constellations of genes and chromosomes brought about by natural selection. In *Poa pratensis* formation of haploids or triploids from predominantly apomictic types also leads to pure sexuality or an increased degree of sexuality.

15. The properties of a haploid *pratensis* type and a number of diploid-triploid twin clones of *pratensis* were studied. The twins with high chromosome numbers require more time to reach their full development than the twins with low numbers. In most cases the twins with an increased chromosome number were less productive than the normal twins. Results of a morphological and chemical analysis of the same material are reported. In *Poa alpina* as well as in *P. pratensis* a positive correlation between chromosome number and cell size was established.

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CONTENTS.

ON THE PROGENY OF DIPLOID × TRIPLOID POPULUS TREMULA

WITH SPECIAL REFERENCE TO THE OCCURRENCE OF TETRAPLOIDY

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BY direction of Professor HERMAN NILSSON-EHLE a series of crosses between ordinary diploid *Populus tremula* L. (2n = 38; cf. BLACKBURN and HARRISON, 1924) and triploid gigas forms of the same species (2n = 57) were carried out at the beginning of 1937 at the Institute of Genetics of the University of Lund at Svalöf (cf. NILSSON-EHLE, 1938). Of the aspen types in question the diploid female type, which was used as a mother plant in the cases treated below, originated from Sätra Bruk in the province of Västergötland, while the triploid males represented two different types, one of which belonged to the clone detected by Professor NILSSON-EHLE in 1935 at Lillö on the shore of the Lake Ringsjön in Skåne (NILSSON-EHLE, 1936) and the other to a clone discovered by S. G:SON BLOMQUIST in 1936 at Våle near Söråker in the province of Medelpad in northern Sweden (BLOMQUIST, 1937).

As reported by MÜNTZING (1936), who has subjected the Lillö type to a cytological investigation, the pollen grains of the triploid display a remarkable variation in size and quality as compared to those of the diploids, due to meiotic irregularities. Thus a certain amount of giant grains are formed, which are supposed to represent the unreduced chromosome number of 57. Providing pollen grains of this type should function, a cross between diploid and triploid would give tetraploid plants (2n = 76) among its progeny. The superior morphological properties of triploid aspens (cf. NILSSON-EHLE, 1938) as compared to diploids, which will enhance their value for industrial purposes, make the breeding of new types of this kind particularly desirable. The production of tetraploids would imply the possibility of securing any number of triploids by crosses with diploids and that was the chief purpose of the breeding experiments in question (cf. NILSSON-EHLE, 1938 and MÜNTZING, 1936).

The crosses were carried out in a greenhouse, where branches of

the above-mentioned types were kept in pots with water.   The catkins of male and female twigs were made to develop simultaneously by retarding the development of the more advanced ones by exposing them for some time to lower temperatures.   When the proper stage was reached, the pollen, which was shed in abundance, was transferred to the stigmata of the female catkins.   Seeds developed within a very short time and germinated after a few days.

At the request of Professor NILSSON-EHLE the seedlings thus obtained were subjected to a cytological examination in order to determine their chromosome numbers, attention being especially directed to the possibility of finding tetraploids.   This task was preceded by a sorting of the material according to the different degrees of vigour displayed by the seedlings.   According to the directions given, the would-be tetraploids, which were supposed to show to an amplified degree the morphological properties of the triploids known, were to be sought for among the most vigorous plants.   Those were all placed at the top of the batch, followed in a descending scale by seedlings of a weaker habit. The plants were numbered from 1 upwards.

The cytological investigation was started as soon as the seedlings yielded root-tips suitable for fixation.   As fixative, diluted chrome acetic formalin was used, and the sections were stained with gentian violet.

It proved very difficult to find really good metaphases — if any divisions were found at all — in which the chromosomes could be counted with accuracy.   Even in the best of plates the exact determination of the number was a difficult task, as the chromosomes, on a whole very small, represent various categories of size, some being very small, others larger, and one individual of each set strikingly large in comparison with the rest.   Consequently, it is sometimes very difficult to distinguish between a couple of smaller chromosomes lying together and an individual of one of the larger size classes (cf. MÜNTZING, 1936). As a rule three counts were made from each plant.   For the above-mentioned reasons the numbers obtained were very seldom the same for all counts, there being usually a difference of a few chromosomes. Sometimes several counts had to be made in order to get a fairly exact value.   On later occasions fixations were carried out at different times and under different conditions, and some of them gave more satisfactory results, the cell divisions being more numerous and the plates fairly good.   As found by TOMETORP (1937), RUNQUIST and the present writer to be the case also with leaf stalks and growing points it proved however difficult to find the most suitable occasions for the cell divisions, which

TABLE 1. *Chromosome numbers of seedlings of Populus tremula from the progeny of diploid × triploid.*

Cross combination	Chromosome number																					n
	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	76	
Sätra × Lillö	3	6	0	3	3	3	4	2	6	8	9	7	11	9	6	3	4	2	0	1	1	91

seem to be much more dependent on external conditions in the aspen than in many other plants. Further attempts will be made in future.
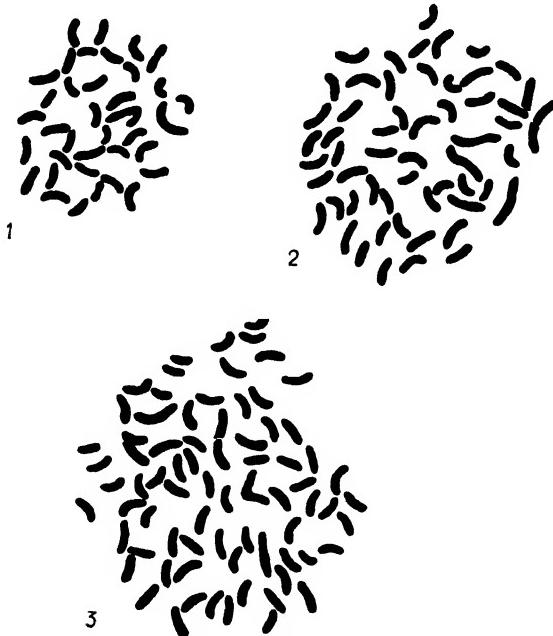
Figs. 1—3. Somatic plates of diploid, triploid and tetraploid seedlings of *Populus tremula* from the progeny of diploid × triploid. — Fig. 1, 2n = 38; Fig. 2, 2n = 57; Fig. 3, 2n = 76. — × 3300. (What seems to be one of the bigger chromosomes in Fig. 3 should be interpreted as two smaller ones lying end to end. In the original drawing this was quite obvious but during the course of reproduction it has become indistinct.)

The investigation was started with the above-mentioned more vigorous seedlings, but also individuals from the remainder of the batch were included as soon as they developed root-tips suitable for fixation; thus an idea of the material in its entirety was obtained. Table 1 shows the distribution of the chromosome numbers among the seedlings of the Sätra × Lillö offspring (PI) examined in the investigation, and

amounted to about 100 plants. The result does not seem to be entirely in accordance with what might be expected from a progeny of this type, where the majority of plants should have chromosome numbers approximate to the diploid value (MÜNTZING, 1936). On the contrary, the values of most seedlings were aneuploid approaching the triploid side. There were only a few plants from which the exact diploid num-

Fig. 4. Approximately triploid giant seedling (2n = ± 55) from the progeny of diploid × triploid *Populus tremula*.

ber of 38 was obtained (Fig. 1, PI—1); some had approximately this number with 39—41 chromosomes, and plants were found representing almost all numbers from 38 to 57. Only one plant was found (PI—169) which seemed to have the exact triploid number of 57 (Fig. 2). Of these plants the diploids and those approximately diploid proved to be rather vigorous plants of a normal development. This was also the case with those approaching the triploid number, but these plants were

distinguished by their conspicuously increased vigour, in some cases resulting in real gigas types with remarkably large leaves and very tall growth (Fig. 4). As a rule the aneuploids were very poorly developed and of a highly divergent appearance. Many of them died at an early stage.

The hope of obtaining tetraploids was fulfilled in that one plant (PI—30) was discovered in the investigation, which was found to have the 2n value of 76 (Fig. 3). Counts were made at first from three roots

Fig. 5. Tetraploid seedling (in the centre) (2n = 76) obtained from the progeny of diploid × triploid *Populus tremula*

which gave fairly good metaphase plates, one or two of which seemed to give the number of 76, while in others only lower values were obtained, but all of them above 70. Re-fixations were made for control and had to be repeated owing to the scarcity of plates. The good fixations obtained confirmed the results of the preliminary counts, giving values varying between about 70 and what seemed to be the exact number of 76, the latter being found in several plates. As is usually the case with plates with a high number of chromosomes and especially with the aspen where, as mentioned above, the difficulties of counting are considerable even at low numbers, there were almost one or two weak

points, however, where it was very difficult to tell with absolute certainty whether one or two chromosomes were present. There·seems, however, to be sufficient evidence to permit the conclusion being drawn that this plant is really a tetraploid.
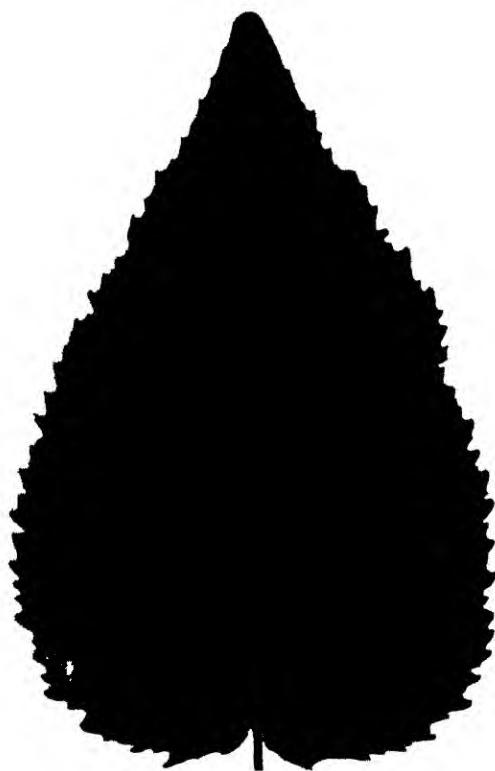
Fig. 6. Leaf of diploid *Populus tremula* (natural size).

As soon as this tetraploid was found the rest of the material was thoroughly searched for plants of a similar appearance, and a few were selected for chromosomal examination, but none turned out to be tetraploid.
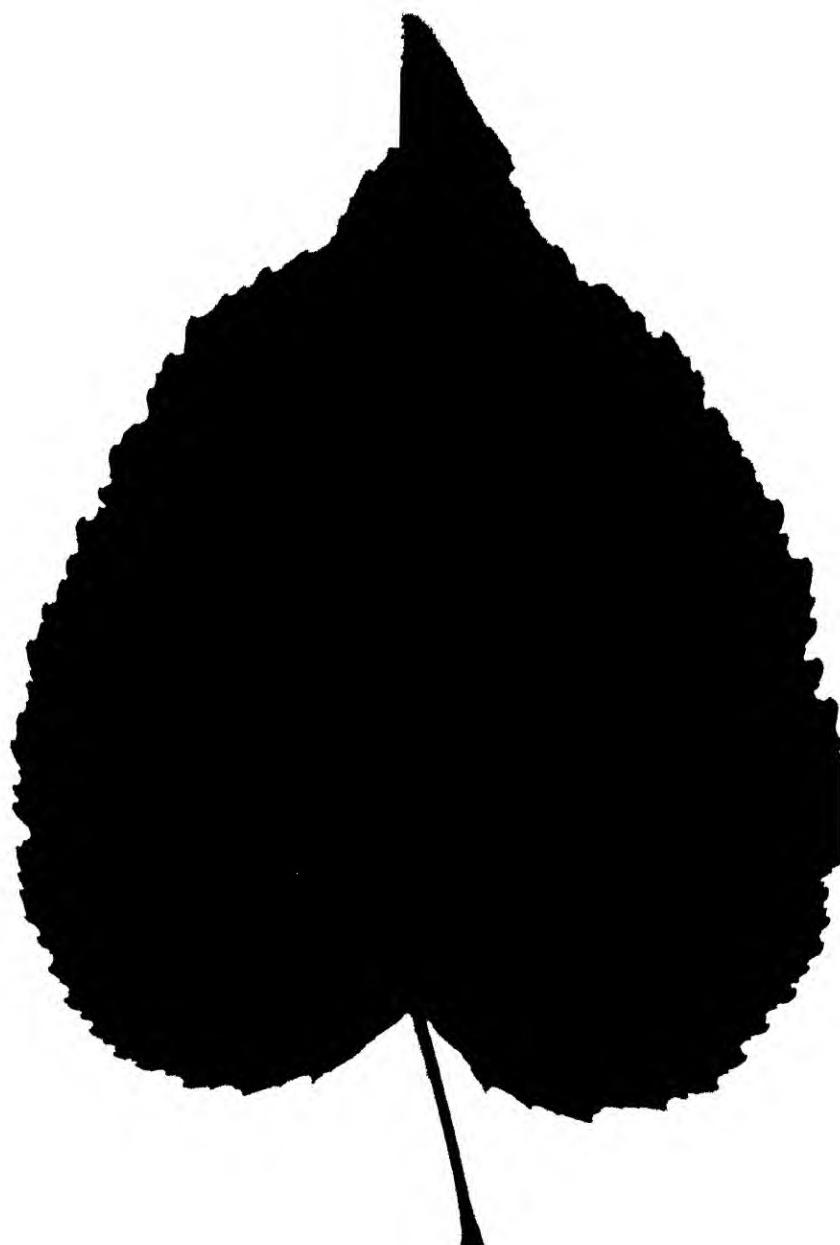
Fig 7 Leaf of tetraploid *Populus tremula* (natural size)

The tetraploid plant (Fig. 5) carried the number of 30 given in the above-mentioned sorting and thus came high on the scale of vigour, though not on the top of it.   Also, it did not at first attract attention owing to any pronounced properties as compared with others, but, as the vegetation period advanced, it developed into a harmoniously built plant of remarkably vigorous growth and had larger leaves than any other plant.   In height it was, however, surpassed by some of the approximately triploid seedlings of gigas type.   Fig. 6 shows a leaf from a diploid plant, and Fig. 7 one from the tetraploid plant.   They

Fig. 8.   Diagram showing the average vigour of seedlings belonging to different classes of chromosome numbers.

both represent the ninth leaf of the plant counting from the base (natural size).

When kept for some time in the beds after having been removed from the greenhouse, the seedlings were subjected to a classification with respect to their more or less vigorous appearance.   By means of ocular gradation they were referred to classes from 5 to 1 according to the higher or lower degree of vigour they displayed.   The results obtained were placed in relation to the chromosome numbers of the seedlings involved.   The diagram (Fig. 8), which shows the average vigour for the different classes of chromosome numbers, gives two maxima, the first at 3,36 for the diploid plants and for those nearly diploid, and the second at 4,80 for those close to the triploid value.   The

minimum occurs at 2,20 for the aneuploids of the chromosomal class 42—46, which contains plants of low viability as also does the class of 46—50, where the slightly higher mean of 2,48 is obtained. Plants approaching the triploid number, i. e. belonging to the class of 50—54, give the higher value of 3,44. The tetraploid seedling, not having at that time yet attained the degree of vigour displayed later on, gives the value of 4. Though comprising rather a limited number of seedlings, about 60, this gradation nevertheless indicates the tendency exhibited by the progeny.

Measurements of the stomata were also carried out, though only on a rather small scale. The values obtained from the four plants involved, two diploids, one approximately triploid with $2n = \pm 55$, and

TABLE 2. *Length of stomata in diploid, triploid, and tetraploid seedlings of Populus tremula.*

	Length of stomata (units)											n	M ± m
	7	8	9	10	11	12	13	14	15	16	17		
Diploid......................	6	21	38	42	29	9	4	0	1			150	9,72 ± 0,12
Diploid......................	3	12	35	37	34	21	8					150	10,21 ± 0,11
Triploid [1].................		4	11	21	40	36	18	16	3	0	1	150	11,55 ± 0,13
Tetraploid...				8	13	33	31	31	23	6	5	150	13,21 + 0,13

the tetraploid, are shown in Table 2. The two diploids examined gave lower values than the approximately triploid giant, while the tetraploid plant shows the highest mean. The result is quite in conformity with the fact exemplified so many times, that an increase of the cell size is one of the effects of polyploidy. As indicated in another paper (BERGSTRÖM, 1938), this may be useful for a preliminary sorting of the material when further selections are to be made.

Fig. 9 is intended to illustrate the morphological variation among the material as exhibited by the difference in leaf type in various plants. The leaves, which represent the largest size-class of each plant, were chosen with respect to this variation and thus do not all belong to plants, the chromosome numbers of which are known. A few instances may, however, be cited. The second leaf from the right in the first row with the very marked dentition originates from an aneuploid plant (PI—56) with ± 45 chromosomes, the first one in the second line belongs to a

[1] Approximately triploid with $2n = \pm 55$.

giant plant (PI—92) with $\pm$ 55, the one next to it (PI—136) has $\pm$ 41 chromosomes, and, finally, the first leaf of the lower line represents a
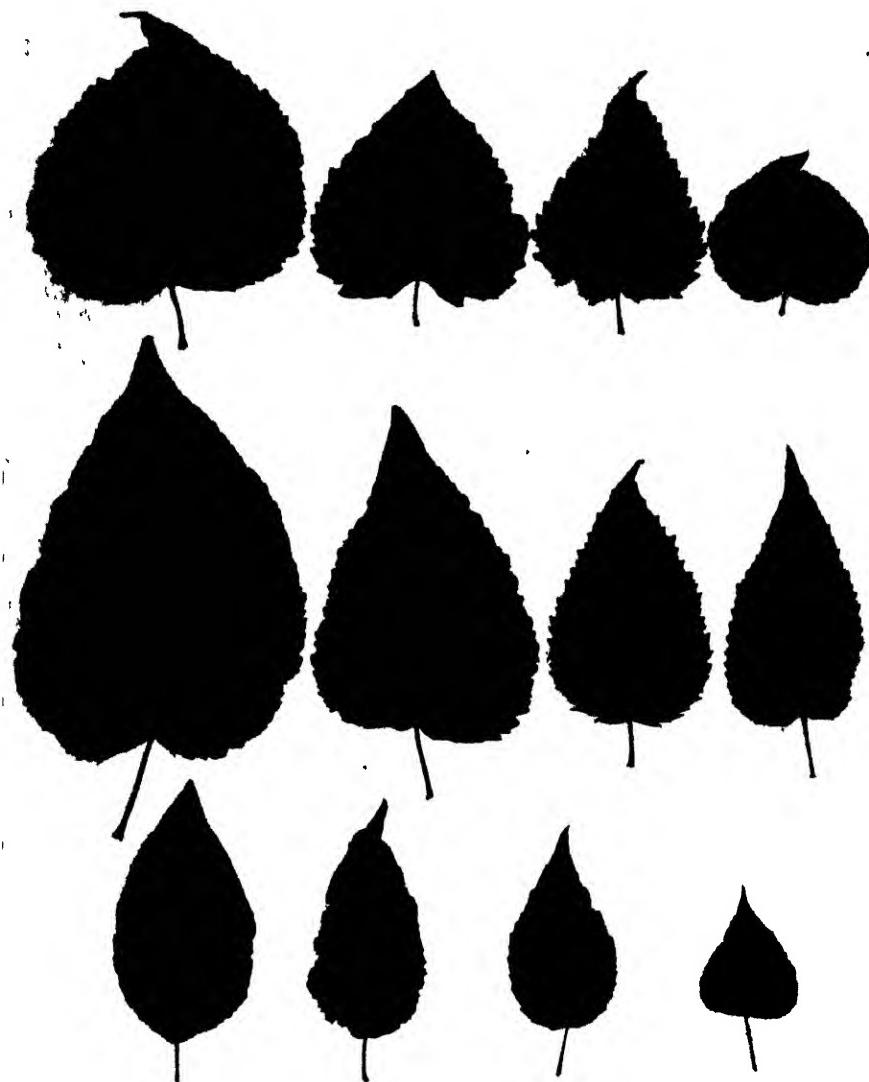
Fig. 9. Leaves of various seedlings from the progeny of diploid $\times$ triploid
Populus tremula.

plant (PI—61) with a 2n value of $\pm$ 48. The length of the leaves is 40 % of their natural size.

Among the seedlings examined from the Sätra × Söråker cross (PII), about 20 only, no tetraploid plant was found. As a whole this material seemed to represent a less favourable combination, the plants being as a rule of rather a poor development.

Since this investigation was carried out a number of new triploid aspens have been discovered at different localities all over Sweden. Some of these new types have been found to be female. There is thus a very extensive material available, on the basis of which further breeding work has to be done and is in fact at present being carried out at the Institute for Breeding Forest Trees at Svalöf.

I am greatly indebted to Professor HERMAN NILSSON-EHLE for having entrusted the carrying out of this investigation to me. I also wish to thank Professor ARNE MÜNTZING for many valuable suggestions, and I am much obliged to Mr. C. G. VON SYDOW and to Mr. E. RUNQUIST for having assisted me in various ways.

Uppsala, April 1938.

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THE CHROMOSOME CONFIGURATION
CAUSED BY AN INVERTED HYPERPLOID SECTION IN DROSOPHILA MELANOGASTER

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IN the analysis of the 3rd chromosome deficiency *Vein* in *Drosophila melanogaster* (MOHR, 1938) advantage was taken of a translocation in which a section of the left arm of the 3rd chromosome (3L) including the free left end, is translocated to the Y chromosome. This Y; 3 translocation was produced by X-rays by PAINTER and MULLER (1929) and we are indebted to Dr. MULLER for the stock and for permission to utilize the case.

The males which carry this translocation have one normal and one broken 3rd chromosome, the left part of the latter being attached to the Y chromosome. Such males have a complete outfit of 3rd chromosome material and are accordingly viable and fertile.

Our analysis of the salivary chromosomes proved that the case is not as simple as previously assumed and a more detailed account will be presented in another connection. For the present purpose it is sufficient to mention that the translocated section which extends from the free left end (61A) of 3L to 72E in BRIDGES' map (1935), was found to contain an inversion of the 63C—72E region.

It cannot now be decided with certainty whether this inversion occurred simultaneously with the translocation as a result of the radiation or whether it was present beforehand in the radiated fly. Dr. MULLER states in a letter that there is no mention in his records of the presence of inversions in the 3rd chromosome.

A photo of the 3L chromosome configuration from a T(Y; 3) individual is presented in Fig. 1 and a diagram of the same in Diagram 1. In the latter the translocation (black) is denoted 1 2 4 3, 1—2 representing the non-inverted, 4—3 the inverted section. The inversion causes the typical loop formation. The remaining section of the broken chromosome (5—6 in Diag. 1) is seen upwards to the right in synapsis with the corresponding part of the normal chromosome (white in the diagram):

In the selection of this preparation for illustration the question of

the relation of the broken chromosome sections to the Y chromosome is left out of account.

Fig 1   The 3L salivary chromosome configuration from a T(Y, 3) $\widehat{XX}$ female. For explanation see text and Diag 1

When T(Y; 3) males are mated to + females we expect ordinary + daughters and sons in equal numbers (see Diagram 2).   The other
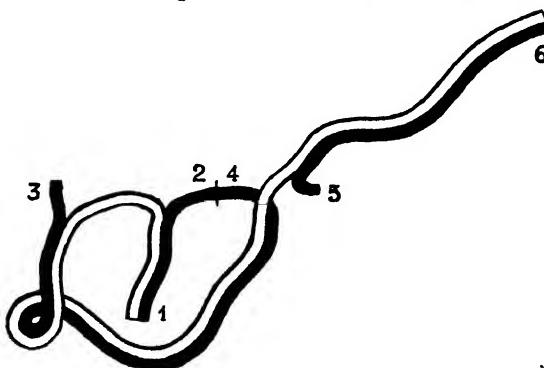
Diag. 1. Diagrammatic representation of the 3L chromosome configuration of Fig 1 Normal chromosome white.   The translocated section (1 2 4 3) and the remainder of 3L (5—6) in solid black.

half of the *male zygotes* will be hyperploid for the translocated section and that such hyperploid males may occasionally survive was observed

by PAINTER and MULLER (1929; MULLER, 1930) who found that they are small and weak, have a blunt body build, a dark patterned thorax and convex wings with imperfect crossveins. They are completely sterile. They are in the following denoted as *dark broad* (*d. b.*).

The actual result of such a test was:

♀♀ + 553; ♂♂ + [T(Y; 3)] 582, *d. b.* 65.

By mating T(Y; 3) ♂ to attached-X ♀ we are also able to obtain

Eggs from:

	✚✚		✚✚
Sperm from T(Y;3) ♂♂	+ ♀	sublethal	+ ♂
	lethal	lethal	lethal
	hyperploid ♂	hyperploid ♀	lethal
	+T(3;Y) ♂	+T(3;Y) ♀	lethal

Diag. 2. The zygotes produced by matings of T(Y; 3) males to + females (left) and to $\widehat{XX}$ females (right). 3L solid black, X and Y stippled.

T(Y; 3) females as well as hyperploid females (see Diag. 2). A certain percentage of the latter survive and were found to show the same somatic characteristics as the hyperploid males.

In this connection it may be mentioned that PAINTER and MULLER in their description of the case (1929; MULLER, 1930) state that the T(Y; 3) translocation is over 25 units long including the loci *ru* (0.0) and *h* (26.5). Our tests demonstrate that the translocated section extends considerably farther to the right, including the *thread* locus (43.2). A

mating of *ru h th st cu sr e* ca* ♀ × T(Y; 3) ♂♂ which in the normal third chromosome carried *ru jv se th st* gave the following offspring:

    ♀♀ *ru th st* 72; ♂♂ + 77, *d. b. st* 7, *ru th st* 1.

The fact that these hyperploid *d. b.* males are non-*ru*, non-*th* but show *st* in spite of the fact that they are known to be homozygous for all these genes, proves that the translocated section contains the normal
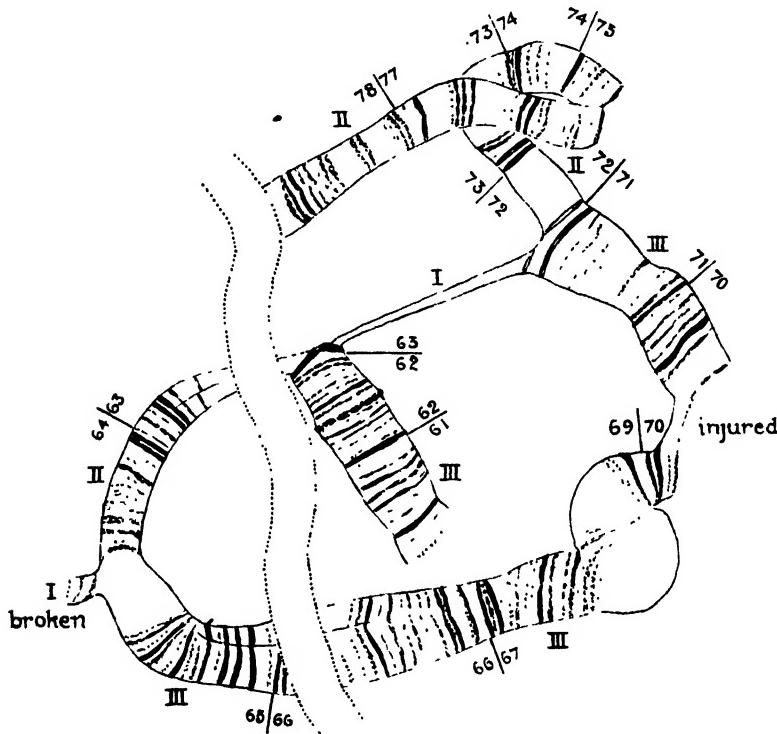
Fig. 2. The 3L salivary chromosome configuration from an X͡X female hyperploid for the 61A—72E section with the 63C—72E region inverted. I = haploid, II = diploid, III = triploid. The distal part of the hyperploid section (to the left) is broken off by accident during the preparation [1].

alleles of *ru* and *th* but not of *st* and that it accordingly ends between *th* (43.2) *and st* (44.0).

The single *ru th st* male obtained in the test is an XO exception due to non-disjunction in the mother.

In the salivary analysis of the translocation crosses of attached-X ♀ by T(Y; 3) ♂♂ were used in order to study the translocation in female

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[1] For the drawings and diagrams we are indebted to the artist of the Anatomical Institute, Miss S. Mørch.

larvae. At the same time this affords an opportunity of studying the synaptic relations in female larvae hyperploid for a chromosome section which contains an inversion.

Preparations from two such hyperploid females have been obtained. Fig. 2 shows the 3L chromosome configuration from such a female. As will be seen the two normal 3L synaptic mates have synapsed along their entire length, and the hyperploid section with the inversion has synapsed with the corresponding part of this diploid chromosome, except for the fact that the distal part of the inverted section has been broken off by accident during the preparation (to the left in Fig. 2). The inversion causes a typical loop formation, principally quite like the loop formed in diploid individuals heterozygous for an inversion.

Fig. 3. Photo of the same preparation as Fig. 2.

Hence the 3L chromosome configuration starts with a triploid section which divides T-like in a diploid (left) and haploid (right) branch. Within the latter. which in this preparation is very much stretched, lies the left end of the inversion. From here on the hyperploid inverted section is in complete synapsis with the corresponding part of the normal diploid chromosome, making the entire loop triploid until the above mentioned broken end. The rest of 3L (from 72E to the spindle fibre attachment) is of course diploid. A photo of the same preparation is presented in Fig. 3.

Fig. 4 shows the 3L chromosome configuration from another hyperploid female larva. Here the critical regions are less stretched and the distal end of the inverted section is complete and connected with nucleolar material. At this point there is an aggregate of chromatic material which probably belongs to (a section of) the Y chromosome, a point which will not be discussed here.

In a diagram of the same chromosome (Diag. 3) the hyperploid translocated section (black) is indicated by the figures 1 2 4 3, 1—2 representing the non-inverted, 4—3 the inverted region as above.

We find accordingly that synapsis is quite regular in the salivary

gland cells of individuals hyperploid for a chromosome section which includes an inversion.

That an inverted section in one haploid chromosome causes the
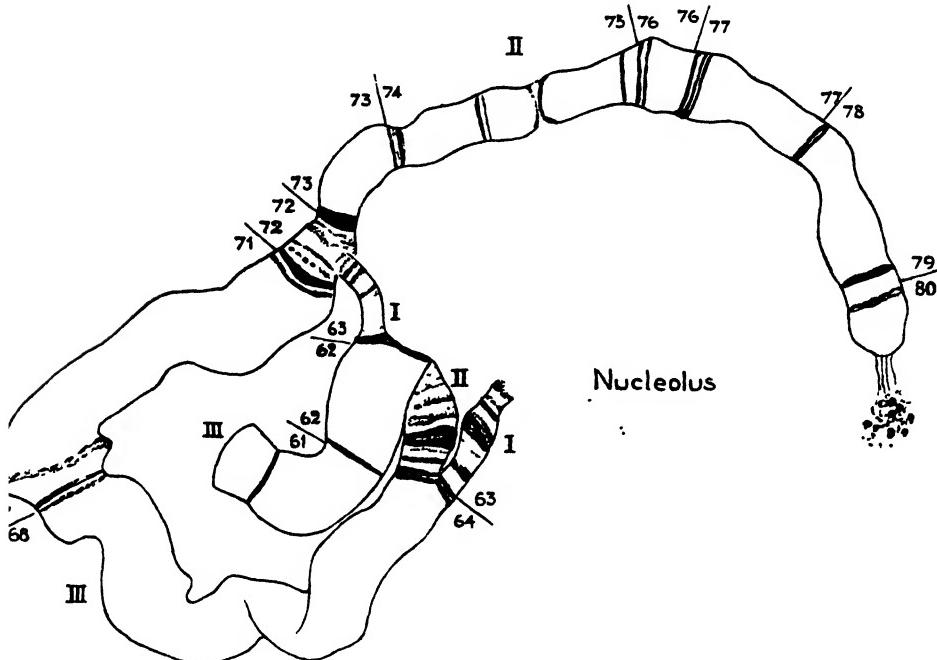
Fig. 4. The 3L salivary chromosome configuration from an X͡X female hyperploid for the 61A—72E region with inversion of the 63C—72E section. I = haploid, II = diploid, III = triploid.
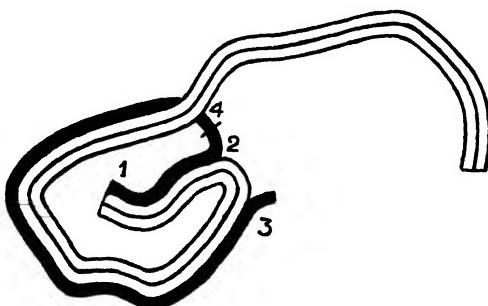
Diag. 3. Diagrammatic representation of the 3L chromosome in Fig. 4. The normal diploid chromosome white, the hyperploid section (1 2 4 3) in solid black.

normal haploid mate to participate in a loop formation during synapsis is quite natural. But it might well be supposed that the presence of an extra chromosome section with an inversion, in addition to the two

normal synaptic mates, might present difficulties which would lead to irregularities.

This is however not the case. The evidence presented demonstrates that there is complete synapsis of the two normal chromosomes resulting in the formation of a normal diploid 3L, and the synaptic forces which cause homologous loci to contact are so potent that an extra haploid section with an inversion forces this diploid chromosome to participate in a loop formation of quite regular type.

SUMMARY.

The salivary analysis of a Y; 3 translocation produced by PAINTER and MULLER by X-rays showed that the translocated section extends from the free left end of 3L, *viz.* 61A to 72E and contains an inversion of the 63C—72E region.

By appropriate matings salivary preparations of female larvae hyperploid for this translocation were secured. It was found that there is complete synapsis of the two normal 3L chromosomes with the hyperploid section, resulting in the formation of a triploid loop corresponding to the inverted region.

Evidence is presented which demonstrates that the translocated section is much longer than previously assumed, extending to a point between *thread* (43.2) and *scarlet* (44.0) in the linkage map.

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THE BALANCE SYSTEM OF MEIOSIS IN HIERACIUM

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INTRODUCTION.

IN this paper the authors do not intend to give a full account of the theory of meiosis, they only wish to take up for discussion some problems hitherto neglected or unknown to cytologists in general. Our opinion is that no morphological findings can explain fully the differentiation of meiosis and mitosis but may nevertheless be of value for a future attack on the causes of meiosis, not only on the course of meiosis. Here we want to emphasize that recent X-ray results have demonstrated quite clearly the time of chromosome splitting in mitosis and the connection between reproduction and nuclear sensitivity. Our interpretation of the facts given below is, however, independent of this view. Similarly we have not discussed things from the view of the precocity theory as outlined by DARLINGTON, although this theory signifies a turning point in the history of cytology.

Recently, one of the authors (GENTSCHEFF, 1937) found a peculiar mode of tapetum development in several apomicts of *Hieracium*. Since pollen production in most apomictic *Archieracium* biotypes is very low or nil, and tapetum degeneration differs from the behaviour in sexual types, there is presumably some causal connection. We began this study with the intention of re-checking the behaviour of tapetum in different apomicts and in pollen-sacs showing different types of division.

Some years ago the other author (GUSTAFSSON, 1935) studied the female development of several apomictic genera and found growth and timing phenomena to be closely connected with changes in meiosis. In a paper published in 1939 a series of apomictic phenomena were shown to depict the same regularity. The occurrence of some exterior factor, changing meiosis and connected with these growth phenomena, was postulated. The other problem to be studied in this investigation was therefore: Do we find in PMC:s with disturbed meiosis growth phenomena or altered time relations responsible for the mitotic-like behaviour?

Finally: What bearing have these eventual growth or time phen-
omena and the balance between tapetum and PMC:s on the explanation
of meiosis?

MATERIAL AND TERMINOLOGY.

In the late spring of 1939 the authors started a series of injection
experiments with folliculin, testosteron, aneurin, auxins and colchicine
on different species of *Lilium*, *Crepis* and *Hieracium* in Lund Botanical
Garden in order to study the hormonal influence on meiosis. For the
sake of comparison untreated plants were fixed. Later we found a
method for the cultivation of *Spinacia* and *Pisum* biotypes under light-
and dark-conditions from seed to flower on agar containing different
salts and hormones, a report of which will be given shortly. For the
original purpose of comparison but also for studies on normal meiosis
we fixed material of several *Hieracium* types. Buds were fixed in the
chromic acid-fixative used at Svalöf Chromosome Laboratory with
some minutes' prefixation in alcohol. The types studied in this paper
are *H. speciosum* HORNEM. and *leiophanum* DT. (2n = 18), *H. caeru-
laceum* ARV. (2n = 27), *H. robustum* MARTR. and *amplexicaule* L.
(2n = 36). A careful examination was made of the two last-mentioned
apomicts. The injection material has not been studied so far, but
apparently the injections cause a series of non-specific artefacts. The
cultivation method mentioned above should be superior.

A great many facts have accumulated from the studies on apomictic
problems during the two last decades. Unfortunately, however, many
scientists working in this field have not denied themselves the pleasure
of introducing new terms or of changing the meaning of old ones. A
most simple but clear terminology should be applied.

In order to facilitate the reading of this paper and to avoid mistakes
we want to give short definitions of the terms and expressions used.

Agamospermy (TÄCKHOLM, 1922): Seed production without fertili-
zation. Comprises the three phenomena of diplospory, apospory and
nucellar embryony.

Allogenomatic and *autogenomatic* (LEVAN, 1937): The genomes are
respectively structurally different or identical. Used by LEVAN for di-
ploid organisms exclusively. The terms can be also used for polyploid
organisms. An autogenomatic tetraploid has four genomes identical
from a pairing point of view, an allogenomatic tetraploid has four

genomes which are structurally different or non-homologous. The wider usage of the terms has been approved of by LEVAN.

Apogamety (RENNER, 1916): The formation of a sporophyte without fertilization from a vegetative cell in a gametophyte.

Apomixis (WINKLER, 1908): Propagation without fertilization.

Apospory (BOWER, 1885): The formation of a gametophyte from vegetative cells of a sporophyte by mitotic divisions. Transitional stages to diplospory exist in phanerogams.

Chromoplasm (KOLTZOFF, 1938): A collective term for the substances, nucleic acids and proteins, which form the chromosome cover or calymma at the kinetic phase of meiosis and mitosis.

Diplospory (EDMAN, 1931): The formation of an unreduced gametophyte from generative cells by means of divisions having meiotic or sometimes a mitotic character. The unreduced chromosome number may arise by the formation of restitution nuclei, pseudohomeotypic divisions or mitotic-like divisions after growth and vacuolisation phenomena.

Hieracium boreale, laevigatum and pseudoillyricum types (ROSENBERG, 1927): The *H. boreale* type has a variable pairing of from no bivalents to many. The *H. laevigatum* type is the extreme case having still contracted, meiosis-like chromosomes but without any pairing. In the *H. pseudoillyricum* type a nuclear contraction takes place at prophase and chromosomes are entirely mitotic-like at later stages.

Interphase (LUNDEGARDH, 1912): The so-called resting stage between two nuclear divisions.

Interkinesis (GREGOIRE, 1905): The transitional stage between first and second division of meiosis. Interkinesis may be more or less a real interphase.

Nucellar embryony: The formation of a sporophyte from nucellar cells without gametophyte formation.

Parthenogenesis (WINKLER, 1908): The formation of a sporophyte from an egg-cell, whether this has arisen in haploid, diplosporous or aposporous gametophytes.

Pseudohomeotypic division (GUSTAFSSON, 1935): No chromosome pairing. Univalents gather in the equatorial plane and divide at first division. No second division occurs.

Semiheterotypic division (ROSENBERG, 1927): No pairing. Univalents are scattered over the metaphase spindle. Restitution nuclei frequently arise. Second division occurs.

Tapetum: One-, two-, four- and eight-nuclear tapetum implies that

the individual tapetal cells contain 1, 2, 4, 8 nuclei. Four-fused tapetum signifies that tapetal cells contain 4 more or less fused nuclei.

Figs 1—24 Meiotic phenomena in *Hieracium robustum*. — Figs 1—3 Division type 1 Meiosis starting at a very early stage — 1. Interphase or early prophase Cells and nuclei very small Tapetum cells one-nuclear. — 2. Semiheterotypic metaphases with crowded, contracted chromosomes — 3. Interkinesis. Micronuclei due to irregular divisions Tapetum cells have one, two or four nuclei. — × 950

DOUBLE CHROMOSOME REPRODUCTION, FRAGMENTA-
TION AND TAPETUM DEVELOPMENT IN HIERACIUM
ROBUSTUM.

H. robustum, never producing pollen, exhibits three types of division in the PMC:s, all connected in some way with the tapetum development, time of division or with the situation of the flowers within a head. In a paper of 1927, ROSENBERG (1927 a) described one regularity with regard to bivalent formation. In biotypes, usually without bivalent formation, rare bivalents occurred, always at the periphery of the head in the very old flowers. This finding was confirmed by GENTSCHEFF (1937) and is true also of the apomicts examined here. Apart from that regularity, ROSENBERG did not discover any conspicuous exterior influences, connected with differences in division type, in spite of the fact that they are sometimes obvious.

H. robustum differs in all division types with respect to tapetum development and division start from the normal as illustrated by GENTSCHEFF (1937). In general early prophase stages of *Archieracium* apomicts appear at the time of one- or two-nuclear tapetum, metaphase and interkinesis at two- and four-nuclear stage. Just a little later the fusion of the four nuclei takes place. Eight-nuclear tapetal cells may also occur.

Division type 1 is found in young flowers in the middle of the head (Figs. 1—3). Cells and nuclei are extremely small (Table 1), the tapetum at the prophase stage still being one-nuclear. Division in PMC:s proceeds rapidly. At metaphase stage chromosomes are contracted and form a semiheterotypic division without bivalents and with the univalents scattered over the spindle. Yet the tapetum cells are in the one- to two-nuclear stage, this being the case at first telophase also. Anaphases are irregular, leading to the origin of polynuclear PMC:s. Regular dyads and restitution nuclei are scarce, an important point in the interpretation of the next division type. In some cases metaphases contain mitotic-like chromosomes or the first division is more or less pseudohomeotypic in character, with many chromosomes arranged in the equatorial plane and splitting lengthwise.

Division type 2 represents a double reproduction of the chromosomes. As is well-known, special tissues of some plants give a continuous increase in chromosome number, due to internal reproductions (GENTCHEFF and GUSTAFSSON, 1939 a and b). The same result was obtained experimentally in *Allium* after auxin-treatment (LEVAN, 1939).

In 1927 ROSENBERG discovered a case in the PMC:s of *Hieracium umbell-*
atum f. apomicta, which seems to be similar, but he interpreted the
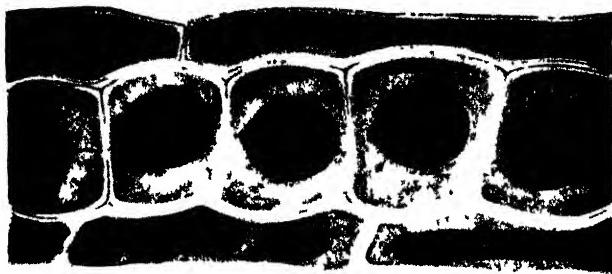
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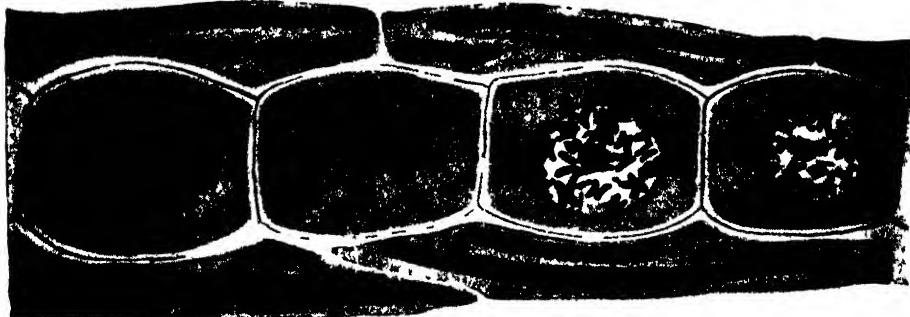
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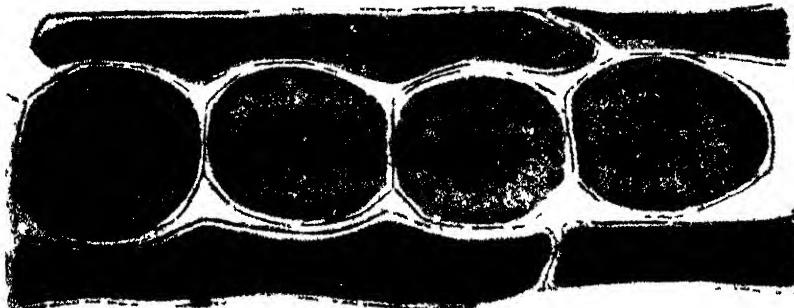
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Figs. 4—6   Division type 2 and 3 — 4   Interphase of division type 2   Tapetum
with four single or fused nuclei. Cells and nuclei have grown intensely — 5 Later
stages with double reproduction (36_{II}, each chromosome apparently having two
chromatids). At metaphase the paired condition is frequently omitted.   Tapetum
very old. — 6 Division type 3   Tapetum cells even larger and older than in 5, but
PMC:s have not grown so much. Owing to the stretching of the pollen-sac tissue
the PMC:s lie separate and have rounded off. — $\times$ 950.

peculiar behaviour of chromosomes and nuclei as being due to the formation of restitution nuclei. After considering all facts and interpretations possible, we conclude that in *H. robustum* another example of the *Spinacia* case is met with. Since chromosome behaviour is different at meiosis and mitosis, the double reproduction will eventually cause an extremely interesting change in meiosis.

In most of the pollen-sacs cells and nuclei continue growing without any divisions beginning. Interphase and early prophase stages — prior to the visible differentiation of chromosomes — occur when the tapetum cells contain two and four nuclei (Fig. 4). Mid- and late prophase are simultaneous with tapetum cells having four single or fused nuclei (Figs. 5, 9). At early prophase PMC-nuclei can sometimes be seen, strikingly resembling mitotic stages from the periblem in *Spinacia* (Fig. 9). Chromosomes appear as long slender threads, lying in pairs and twisted around each other like chromatids (relational coiling). Whether the chromosomes themselves consist of two chromatids or not cannot be decided with certainty; in suitable places they appear double, similar to the case in *Spinacia*. In more advanced stages the chromosomes contract but are still coiled once or twice around each other and lying in pairs (36_{II}). At the same time as they decrease in length, chromosomes become denser and thicker and their breadth is definitely greater than at interkinesis or early second prophase (Figs. 7, 8, 12, 13). At late prophase the mutual coiling has disappeared but most of the chromosomes are in pairs. Frequently median connections are seen. Presumably they are due to a delayed centromere division as in auxin-treated *Allium* species. After the end of prophase — more or less diakinesis-like — the chromosomes have contracted even more, and at metaphase they exist as large round elements (Figs. 17, 18). Pairs of chromosomes still occur in many nuclei. Frequently, however, the individual chromosomes have separated and form a typical semi-heterotypic division, the univalents being scattered all over the spindle. Sometimes metaphases with long and slender, mitotic-like chromosomes are found.

In the same pollen-sacs where this double-reproduction has taken place, cells with the single chromosome number (36) also occur (Figs. 10, 14, 15). The number of these tetraploid PMC:s is variable. In no case has bivalent formation been observed in the median flowers. Apparently bivalents cannot arise in sacs with double reproduction, the nuclei either containing the double or the single number.

Prophase chromosomes of these tetraploid nuclei present the com-

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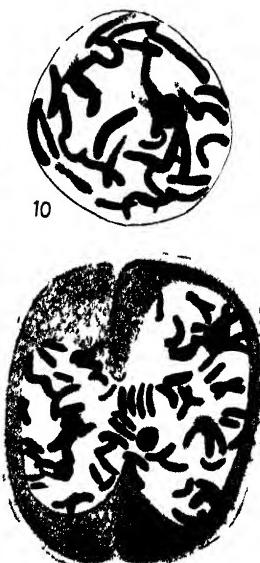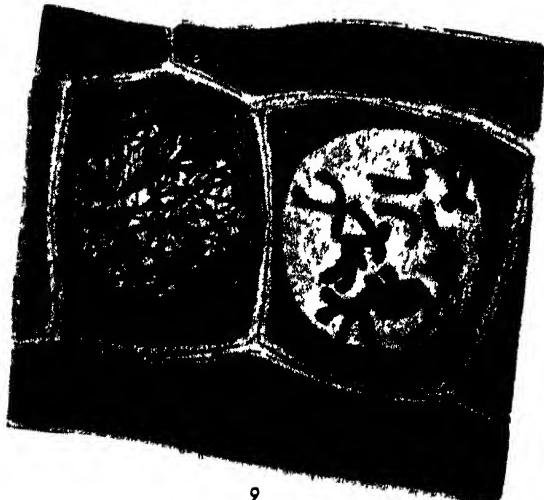
mon appearance, characteristic of *Hieracium*-apomicts having the *boreale*- or *laevigatum*-types of meiosis. They are fairly long and thick, always with the two chromatids closely attracted. At metaphase they are strongly condensed, almost round in shape. Generally forming a semiheterotypic division they lie scattered over the spindle. Sometimes they enter the equatorial plane and give a pseudohomeotypic division, usually irregular. Save for the chromosome number octoploid and tetraploid metaphases with scattered univalents appear completely identical. Anaphase separation is disturbed in both types of cells, and interkinesis-stages with micronuclei are common. The study of second division is rendered difficult, because PMC:s start to degenerate already before metaphase. In some heads and flowers the early degeneration is especially pronounced. In those very aberrant cases tapetum cells differ markedly from the normal appearance. They are swollen and contain vacuole-like formations. Usually the chromatin of the tapetum is discoloured or has begun to dissolve. In many flowers degeneration of the PMC:s is not expressed until the first telophase or interkinesis. At any time when PMC:s degenerate, tapetum cells are abnormal and have reached the 8-nuclear or 4-fused stage. The correlation is quite clear.

The premeiotic interphase and the early meiotic prophase are extremely mitotic-like; in fact, with the exception of differences in size the similarity between tapetal and PMC-nuclei is conspicuous. As has been mentioned already, early prophases of double-reproduction nuclei are remarkably reminiscent of periblem divisions in *Spinacia*. This state of things must be kept in mind in any discussion of the results.

As mentioned above, ROSENBERG (1927 b) explained the occurrence of similar pictures in *H. umbellatum* f. apomicta by assuming the formation of restitution nuclei. In some microsporangia of this apomict the PMC:s were old and rounded off before division set in. In such divisions bivalents formed frequently. At first telophase two nuclei arose, often very different in size. Small cells remained undivided,

Figs. 7—10. Divisions in pollen-sacs with double reproduction. — 7 and 8. Two PMC:s cut into two portions, each of them with 36 pairs of chromosomes. In some cases the individual chromosomes have become free, in others they lie close, due to a previous relational coiling (the twisting is still visible). — 9. One very early prophase with double reproduction, similar to periblem-divisions in *Spinacia*. The relational coiling is apparent. In the PMC to the right prophase is more advanced. — 10. Semi-heterotypic prophase (36$_1$) from a micro-sporangium with double reproduction. The nucleus is small. — 11. An interkinesis-stage (2nd prophase). Note the smaller size of chromosomes, nucleus and cell as compared with Figs. 7—9. — $\times$ 2100.

their nuclei being at an interphase-stage. In some cells the nucleus is in an interkinesis-like prophase with split chromosomes. In the common type of restitution nuclei there are ·27 split chromosomes, but in these cases both single and double chromosomes were present. The explanation of the high chromosome number is, according to ROSEN-BERG, that anaphases of PMC:s with a previous bivalent formation
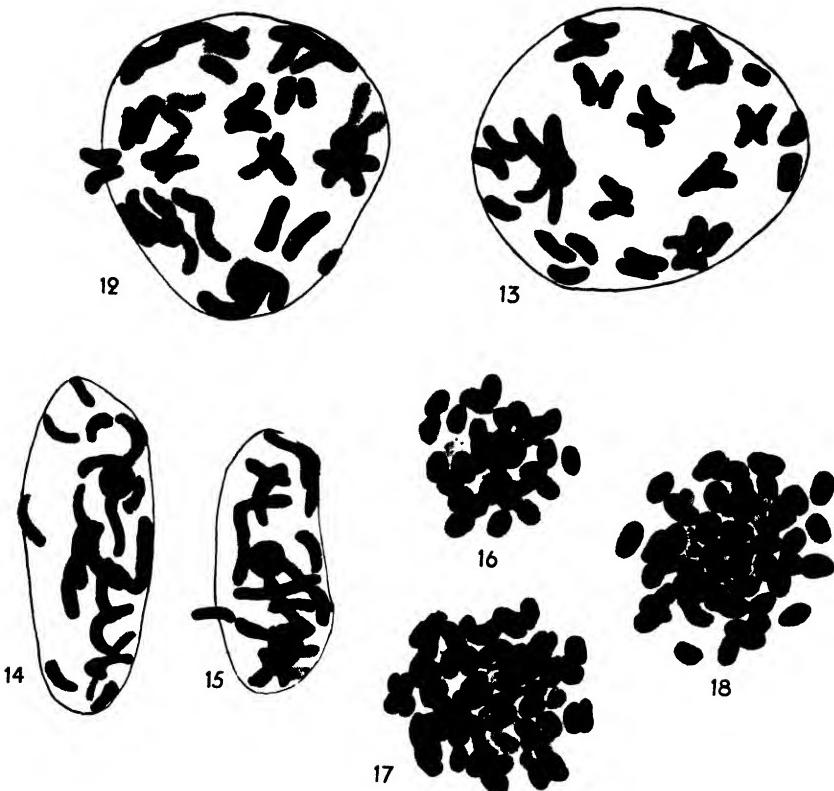
Figs. 12—18. Division type 2 continued. — 12 and 13. Two portions of the same PMC (36_{II}, fragments are due to the sectioning). — 14—15. One prophase nucleus with 36 univalents (actually 38 bodies occur, the higher number is due to the sectioning). — 16—18. Metaphases from cells with the tetraploid number (Fig. 16), from cells with 36 pairs of chromosomes (Fig. 17) and 72 univalents (Fig. 18). In all metaphases chromosome size and structure are identical. — × 2100.

obtain disjoined bivalent chromosomes, each consisting of two chromatids. Univalents gather at the equator and divide already at first division. Then a nuclear membrane is formed around all chromosomes. During the subsequent interphase (interkinesis) those chromosomes formerly participating in bivalent formation split. The chromatids of these chromosomes lie in the form of pairs and at second

metaphase they move to the equator. The chromatids of the univalent chromosomes do not split at second division, being the result of a splitting in first division, and so they remain at the outskirts of the spindle. Whether ROSENBERG's interpretation is true or not, we do not venture to decide. Some properties of these supposed restitution nuclei are, however, suspicious. All the nuclei examined contained exactly 54 chromosomes (2×27), no more, no less. As restitution nuclei are often incomplete, lower numbers could be also expected. The supposed interkinesis-chromosomes differed in shape and appearance from the normal, and metaphase chromosomes were contracted as at first metaphase. This last feature is especially surprising. Taking all these facts into account, we cannot disregard the possibility of a double reproduction even in *H. umbellatum* f. apomicta. In such a case the single chromosomes of a pair would have slipped off each other rather early. In fact, at least two of the singles in ROSENBERG's Fig. 2 A lie close, and two others somewhat more apart. The same is true also of Fig. 2 B. In the double reproduction nuclei of *H. robustum* we have often found that the separation of paired chromosomes may begin prior to the disappearance of nucleolus and nuclear membrane.

By various means it can be proved that the double reproduction in *H. robustum* cannot be explained in the manner outlined by ROSENBERG for *H. umbellatum*.

1) The earliest divisions visible (belonging to type 1) are very irregular. Bivalents do not arise. Restitution nuclei are rare and, if formed, often incomplete. Bivalents have been seen but exclusively in the peripheral flowers. The corresponding divisions begin even later than those of division type 2. Bivalent formation is not prior to but later than double reproduction. — 2) In the event of double reproduction the whole of a pollen-sac contains cells with one single nucleus (in contrast to the conditions in ROSENBREG's investigation). If these single nuclei were restitution nuclei, two- or poly-nuclear cells should be found side by side. — 3) The interphase is more similar to the corresponding mitotic stage than to an interkinesis. In fact, the granular or net-like structure of PMC:s and tapetal cells is identical. — 4) These interphases give rise to prophases with normal unpaired chromosomes (36) close to the double reproduction nuclei. Corresponding stages are seen in both types of cells. The prophase chromosomes of doubled nuclei are no doubt different from those of interkinesis. They are thicker and broader with a ratio of length: breadth much smaller than in mitosis or second division. The chromosome number is always either 36_I or

36_{II}, and only at the end of prophase or transition to metaphase do the chromosomes of a pair separate. — 5) Early prophases look different from interkinesis-stages. The threads are twisted around each other and are longer and more slender than in a second prophase. — 6) Metaphases are similar whether they contain 36_I, 36_{II}, 36_{II}—72_I or 72_I. Chromosomes are extremely contracted, square or round in shape and different from those at second metaphase. They always lie scattered over the spindle or form an irregular pseudohomeotypic division later on. Even in the event of a remaining pairing (this is not a pairing by contact) the division is semiheterotypic, indicating that the centromere of a pair has divided previously, each chromosome possessing at least one centromere of its own.

Summarizing these facts, we must conclude that the occurrence of restitution nuclei after a preceding metaphase cannot explain the origin of double numbers in *H. robustum*. These must be due to definite changes of the reproduction mechanism. Since the earliest prophases visible contain double, twisted threads, as in *Spinacia*, most probably the double reproduction occurs somewhere during the interphase where PMC:s and nuclei grow intensely (Table 1). In *Hieracium*- and *Taraxacum*-apomicts, of either the male or female organs, due to the omitted chromosome pairing, no typical pachytene stages occur. The transitional stage of interphase and typical prophase is sudden. If DARLINGTON's postulates as to the time of splitting of meiotic chromosomes are true — and there is plenty of evidence — we dare not deny the possibility that in these double-reproduction nuclei of *Hieracium* splitting is repeated twice *at early prophase*. It seems hardly plausible, however. Here we wish to emphasize the fact that the interphases in division type 2 are extremely mitotic-like.

Division type 3 is associated with bivalent and fragmentation phenomena (Figs. 6, 19—24). As mentioned above, bivalents do not arise in the median flowers of a head, they are restricted in origin to the peripheral microsporangia. The cause of this will be discussed later. Prophases and metaphases of division type 3 occur at very advanced tapetum stages, even more advanced than in division type 2. Four-fused and eight-nuclear tapetal cells are most common (Fig. 6). The number of bivalents is variable: the occurrence of 9—10 bivalents is the rule, corresponding to an eventual hybrid structure $(9_{II} + 18_I)$. Trivalents occur but not frequently. Cytologically — even in the case of strong pairing — this apomict is not autopolyploid (or autogenomatic, which is a better expression; LEVAN, 1937). At anaphase chromosome

separation is irregular, due to the high number of univalents. Inversion-bridges are frequent, at first as well as at second division (Figs. 22, 23). In the same plate three or four bridges may be found, indicating a high degree of structural differences. At first division the occurrence of
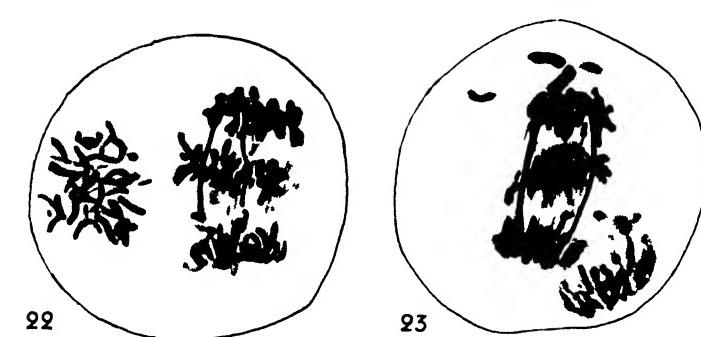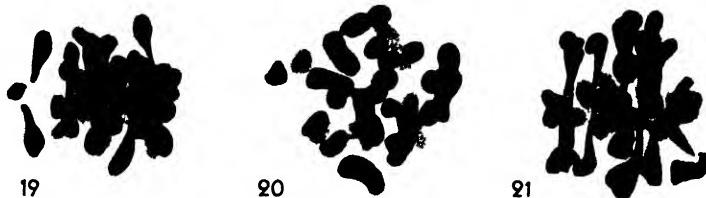
Figs 19—24 Behaviour in the case of bivalent formation — 19—21 ± 10, 9 and ± 11 bivalents respectively — 22 and 23 2nd meta- and anaphases with two-centromere formations due to inversion-bridges Chromatids from univalents, split already at first division, are seen in the equatorial plane — 24 Fragmentation in three PMC s Traces of bivalents and even trivalents can be seen — × 2100

inversion-bridges sometimes gives rise to hour-glass-shaped restitution nuclei of a peculiar shape. At second anaphase chromatids frequently remain in the equatorial plane, demonstrating the splitting of univalents already at first metaphase-anaphase. In spite of the structural differences most chiasmata are terminal, each bivalent generally possessing only one chiasma. Either this indicates an exclusive pairing of terminal segments or, what is more probable, owing to the incidental occurrence of bridges, it indicates that terminalisation proceeds over homologous chromosome parts as well as over sections structurally different.

The most conspicuous feature in the case of bivalent formation is the extreme fragmentation of the chromosomes (Fig. 24). This destruction does not begin until late metaphase, but then the cells are filled with small chromatin pieces of different size. Now and then traces of bivalents (trivalents) can be seen, when the fragmentation was not complete. An interesting point is that fragmentation is higher at the tip of the pollen-sac than close to the ovule. This is a correlation phenomenon. A similar condition is found in *H. amplexicaule*. The fact has not been noticed previously but is very marked.

Similar fragmentation phenomena have been described by BEADLE (1932), BERGMAN (1935), MATHER (1934), and WHITE (1937). In the first case a spontaneous gene-change altered the properties of meiotic chromosomes so that they become sticky and break into pieces. In his paper, MATHER demonstrated a different response of chromosomes to X-rays in *Tradescantia bracteata* and *Vicia faba*. In the former species, where terminalisation is almost complete, fragments arise already at prophase; in the latter species, where chiasmata are interstitial and numerous at metaphase, fragmentation does not begin until early anaphase. (Unfortunately the fragmentation rate of *Vicia faba* was fairly low.) According to MATHER, the different time-action of repulsion forces is responsible for the difference in behaviour. It could possibly be assumed that the formation of bivalents and their separation caused the altered stability of the chromosomes also in *H. robustum*. In that case, however, only the bivalent chromosomes would break and the univalents, which are frequent, would remain intact. This is not true. All chromosomes, whether from bivalents or univalents, finally fall into pieces. Therefore the explanation of the fragmentation in *H. robustum* must be another. In the lateral, very old flowers conditions exist which cause 'bivalent formation and at the same time increase the internal weakness of the chromonemata. Fragmentation in *H. amplexicaule* is

somewhat different, but even there it does not start until anaphase (p. 232). On p. 244 these matters will be discussed further. Here only one interesting fact will be mentioned.

Restitution nuclei and telophases of the first division pass into a real interphase (resting stage) in many *Hieracium* species. Therefore we have at first anaphase-telophase an onset of forces, changing the chromosomes to interphase structure. Presumably the destruction of nucleic acids and proteins, formed at early prophase (CASPERSSON, 1936; KOLTZOFF, 1938) and producing the chromoplasm (matrix, pellicle, calymma), sets in, rendering the chromonemata (genonemata, KOLT-ZOFF) more or less free. As long as the chromosome cover persists, the weakened chromonemata are held together; but when the kinetic phase comes to an end and the non-persistent material of the chromosomes disappears, the weakened chromonemata will release the hundreds or thousands of micro-units.

That the delayed onset of prophase in PMC:s can weaken the chromosomes, is also shown by BERGMAN's paper (1935). In strongly hydrated cells of *Leontodon hispidus* chromosomes were usually mitotic in length at metaphase, but at the same time intensely fragmented.

GROWTH AND TIME PHENOMENA IN H. ROBUSTUM.

In Table 1 the size of PMC:s in different types of division can be seen. The earliest divisions to start (division type 1) lack bivalent formation but chromosomes are contracted (i. e. heterotypic in shape) and the anaphase implies an actual reduction in chromosome number, apart from the few cases of restitution nuclei. Cells and nuclei are very small. The greatest cell-growth takes place between early prophase and metaphase. At metaphase cells have approximately the same size as at interkinesis (products of length and breadth = 156 and 162 respectively). The small size of the cells even at metaphase makes the cells intensely crowded with chromosomes.

Division type 2 shows a very strong growth of the interphase cells before meiosis begins. They are larger than the interkinesis cells of division type 1 (193 : 162 square-units). If they were actually restitution nuclei after semiheterotypic divisions of type 1, their size should not be greater than that of interkinesis cells. In the event of double reproduction the increase in size is very marked at early prophase, whereas prophases with single chromosomes (close to the prophase nuclei with double reproduction) have a slightly higher cell-size than at

interphase (in the first case 403 to 193 square-units, as against 261 to 193 square-units in the latter case). Cells with 36_{II} or 72_I have approximately the same size at metaphase as at late prophase. This difference in behaviour can be explained in the same manner as in *Spinacia* (GENTCHEFF and GUSTAFSSON, 1939 b, p. 384). Due to an intense growth at interphase many cells have acquired a size that does not correspond to the chromosome number. Still the somatic number may not be

TABLE 1. *The PMC-size of H. robustum in different division types.* (*The average figures of ten PMC:s. 1 unit = 1,3 μ*).

	Interphase			Prophase			Metaphase			Interkinesis			2nd Anaphase		
	Length	Breadth	Product	Length	Breadth	Product	Length	Breadth	Product	Length	Breadth	Product	Length	Breadth	Product
Division type 1......		--	--	5,05	14,0	70,7[1]	9,2	17,0	156,4	10,2	16,0	162,4	—	—	—
Division type 2 a) Double reproduction...............	12,9	15,0	193,5	21,2	19,0	402,8[2]	23,2	19,7	457,0	—	—	—	—	—	—
b) Single reproduction...............	—	-	—	15,7	16,6	260,6[2]	--	—	-	-	—	—	—	—	--
Division type 3 a) Bivalent formation...............	—	—	—	—	—	—	16,4	14,8	242,7	—	—	—	19,7	16,3	321,1
b) Fragmentation ...	—	—	—	—	-	—	14,2	18,1	257,0[3]	--	—	—	-	-	—

increased at the time of prophase onset. If, however, the cell-size has advanced beyond a certain threshold value, reproduction must continue until cell-size and nucleus-size are in harmony. As nucleus-size is proportional to the chromosome number, an increase in nucleus-size can be brought about by internal reproductions. After the double reproduction, nuclei will continue to grow until an optimum size has been reached, hence the prophase increase of growth. (Cf. in *Spinacia* the different size of 12_{II} and 24_I cells.) In cells below the threshold value one single reproduction will occur (either at interphase or at very early prophase), hence the slight increase in the size of single-prophase cells from the same pollen-sacs where double reproduction is also found.

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[1] Corresponding to early prophase. — [2] Corresponding to late prophase. — [3] Corresponding to beginning anaphase.

In division type 3 cells with bivalents are larger than interphase cells of type 2. ROSENBERG's explanation, cited above, cannot be true of *H. robustum* even for that reason. If the interphase cells of type 2 contained restitution nuclei formed in cells with bivalents, they should be larger than metaphase cells of type 3 and only slightly smaller than cells in second division (the ratio is here 193 : 321 square-units).

As shown in the table, metaphase cells of type 3 are very much smaller than those of type 2 (proportions 243—257 : 457). That is seen also in Figs. 5 and 6. In spite of the prolongation of interphase and the late onset of division they have grown only a little. Due to the stretching and growth of the pollen-sac tissue they have, however, rounded off. At the same time they show a high rate of bivalent formation — hence meiosis is more normal in this respect than in division types 1 and 2. The double reproduction cannot be due to an inhibited precocity of the cell-divisions but to the growth of the cells. And, what is more important, a long duration of the interphase stage does not inhibit meiosis (cf. GUSTAFSSON, 1939).

Nuclear size has been measured in the same manner (length, breadth and product of ten nuclei). Early prophase nuclei of type 1 show the dimensions 4,3 and 8,2 units (product $= 35,3$). Interphase nuclei of type 2 are 8,7 units long and 9,4 units broad (product $= 81,8$). Double reproduction nuclei at late prophase give the figures 14,6 and 13,1 (product $= 191,3$), single reproduction nuclei from the same microsporangia 11,0 and 10,3 (product $= 113,3$). There is a striking increase in growth at prophase of type 2, if double reproduction has occurred, but not in the case of normal behaviour. Had these interphase nuclei arisen after restitution processes, their size would not change so remarkably at prophase. Interphase nuclei of type 2 are much larger than prophase nuclei of type 1.

SEMIHETEROTYPIC AND PSEUDOHOMEOTYPIC DIVISIONS, BIVALENT FORMATION AND FRAGMENTATION IN HIERACIUM AMPLEXICAULE.

Like *H. robustum*, this apomict is tetraploid ($2n = 36$). Pollen can be produced but is usually poor, probably not viable. In *H. amplexicaule* a different metaphase behaviour appears and a definite regularity with regard to the time occurrence and location of the flower has been found. Most frequently three division types occur, i. e. semihetero-typic division after prophase stages without any so-called synizesis-

phenomena, pseudohomeotypic division and bivalent formation after prophase stages showing synizesis. In this apomict bivalent formation is not associated with any fragmentation of the chromosomes. Instead, this occurs at pseudohomeotypic meta- to anaphase in a special manner. Besides these three division types — which give transitions as described
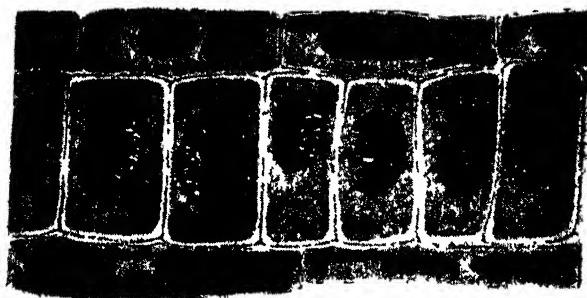
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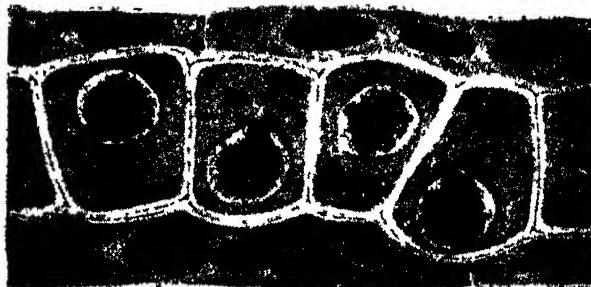
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Figs 25—55. Meiotic phenomena in *Hieracium amplexicaule* — 25 PMC-appearance and tapetum development in prophase type 1, leading to semiheterotypic divisions. — 26. PMC- and tapetum appearance in prophase type 2, leading to pseudohomeotypic divisions and bivalent formation Note the advanced tapetum and the PMC-growth. — × 950

below — some cases of omitted chromosome contraction have been observed and also some cases of an extreme delay of prophase start.

Two common types of prophase nuclei have been observed, one occurring in the median flowers of a head at an early stage of tapetum development (Fig. 25), one in the lateral flowers in late stages of tapetum development (Fig. 26). As mentioned above, the two processes giving rise to different prophase development, usually cause also a different behaviour of the chromosomes at metaphase. A third pro-

phase type occurs after intense nuclear and cellular growth. This third type did not produce metaphases in the material examined.
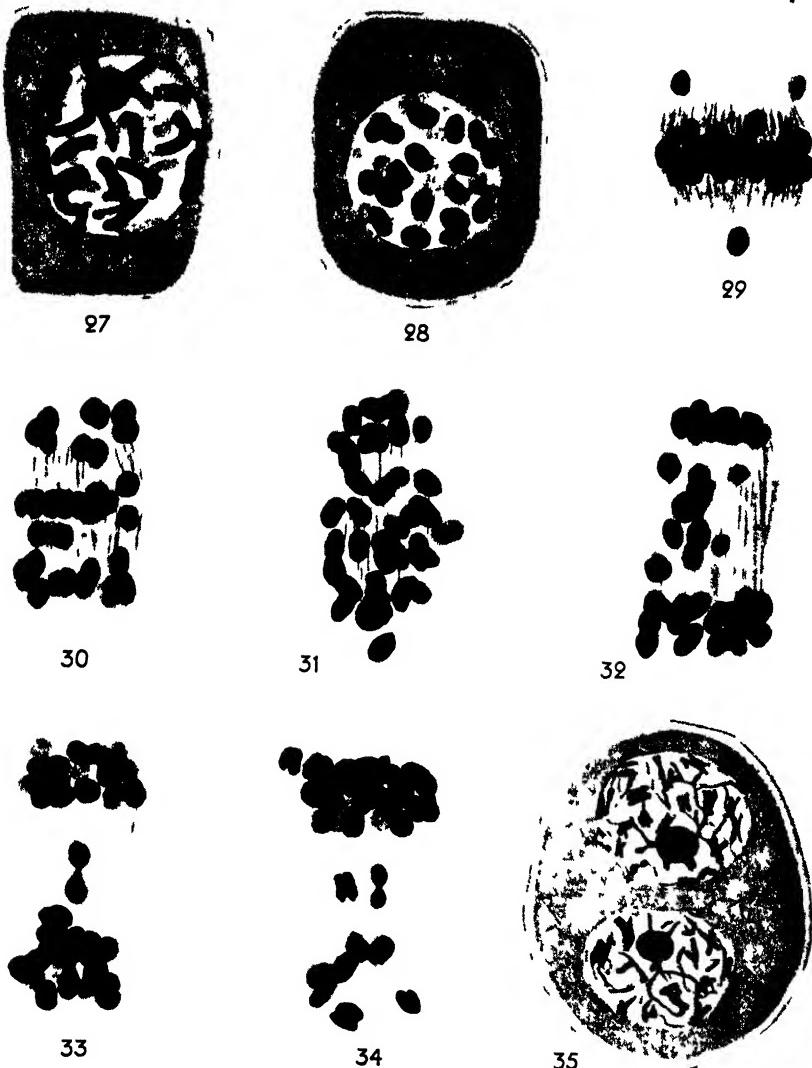
Figs 27—31 Semiheterotypic divisions in the median flowers after prophase type 1 — 27. Early prophase. — 28 Late prophase with contracted chromosomes. — 29 Transition between semiheterotypic and pseudohomeotypic division. Regular pseudohomeotypic metaphases are not formed after prophase type 1. — 30. Semiheterotypic metaphase. — 31. Ditto — 32—34 Atypical semiheterotypic divisions as transitions to the later stages after prophase type 2. — 32. One bivalent-like formation. Anaphase. — 33. Anaphase with a bivalent-like formation. — 34. Two differently splitting univalents. Usually univalents do not split at first division in the case of semiheterotypic anaphase. — 35. Interkinesis. — ✕ 2100.
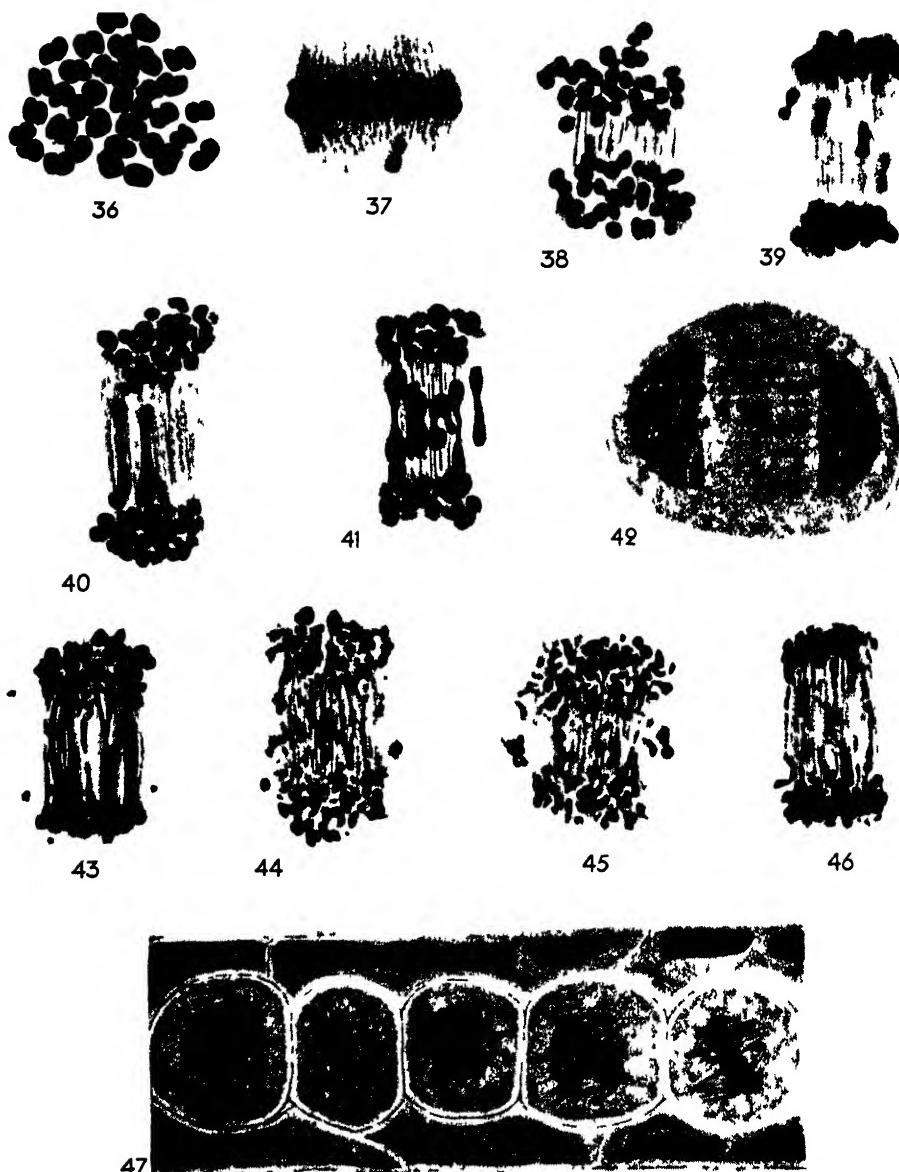
Figs. 36—47. Pseudohomeotypic divisions and fragmentation phenomena —
36. Pseudohomeotypic metaphase   Polar view — 37. Ditto   Side view. —
38. Pseudohomeotypic anaphase. — 39. Ditto with some delayed univalents —
40. Pseudohomeotypic division combined with bivalent formation   Univalents split
before the bivalent separation. — 41. Bivalent formation and some splitting uni-
valents. — 42. Pseudohomeotypic interphase.   Compare the shape of the nuclei
with that in Fig 35 — 43. Inversion-bridges after bivalent formation.   Fragments

Prophase type 1 in *H. amplexicaule* takes place when the tapetum cells contain one or two nuclei. Like *H. robustum,* this apomict has very small PMC:s when meiosis begins. Division starts precociously with respect to tapetal development and cell-growth. The subsequent metaphase contains univalents exclusively; bivalents do not and probably cannot arise (Figs. 27—31). Semiheterotypic metaphases with highly contracted chromosomes are the result of this unpaired condition. At anaphase chromosomes are passively moved to the poles, their distribution being irregular. Restitution nuclei are formed frequently, due to the occurrence of univalents remaining in the middle of the spindle (for the process of restitution, see ROSENBERG, 1927 a and GUSTAFSSON, 1935). Not infrequently, however, some univalents gather in the equatorial plane (apparently by an active movement of their own) and split lengthwise (Fig. 30). If the number of splitting univalents is high at first meta- to anaphase, we get a pseudohomeotypic division. In the median flowers and after prophase type 1 regular pseudohomeotypic divisions are rare. Most frequently we find transitions to the semiheterotypic state. Parenthetically it must be mentioned that in semiheterotypic divisions of most apomicts in *Hieracium* and *Taraxacum* the chromatids of univalents do not as a rule separate from each other until second metaphase, as in sexual species, and only in the event of bivalent formation do some univalents move to the equator and split lengthwise already at first division. In the meta- to anaphases, beginning early, no fragmentation phenomena have been seen. Instead, these are associated with the second prophase type and its later stages.

Prophase stages of type 2 appear when tapetum cells contain four single or fused nuclei and arise in the lateral flowers of a head. Transitions between prophase type 1 and 2 exist. In the narrow zone of half-median (or half-lateral) flowers divisions having another appearance occur. In general, PMC:s containing prophase stages of type 2 have grown larger than in type 1. Therefore they have not begun division until a well-developed tapetum has been formed and cells and nuclei have grown out to certain minimum dimensions.

As in sexual species this prophase type, characterized by a sensitive prophase stage, gives rise to bivalent formation. Even· pseudohomeotypic divisions of almost regular and typical appearance are common

due to the inversions. — 44—46. Intense fragmentation combined with pseudohomeotypic division. — 47. Fragmentation. PMC:s to the right are closer to the ovule. Fragmentation is more intense towards the top of the loculi. — Figs. 36—46, × 2100. Fig. 47, × 950.

(Figs. 36—39). Apparently pseudohomeotypic divisions cannot arise, or semiheterotypic metaphases are exclusively formed in cells where the previous prophase or interphase stages showed no growth phenomena or started precociously. If DARLINGTON's hypothesis regarding chromosome movements is correct, univalents must ripen in order to obtain a two-centromere condition, and not until this process has finished are they capable of movements, orientation and division of their own. This conclusion was drawn by GUSTAFSSON in 1935 with regard to pseudohomeotypic divisions on the female side of *Taraxacum*. Several cases of pseudohomeotypic divisions in male organs have already been described. With regard to their regularity there is a striking difference between the two types of sex-organs (GUSTAFSSON, 1938). In late or outgrown PMC:s of·*H. amplexicaule* typical divisions occur. Even here transitional stages to the scattered position of univalents are sometimes found. They are not so common, though, as after prophase type 1.

Bivalent formation (*H. boreale*-scheme) varies greatly (Figs. 48—52). As few as one or two bivalents occur frequently, but as many as nine (or even more) bivalents may arise. Apparently the maximum number of bivalents is approximately identical to what an eventual hybrid structure would indicate ($9_{II} + 18_I$). *H. amplexicaule*, like *H. robustum*, must be regarded as allogenomatic.

In some PMC:s we have noticed a bivalent behaviour and an anaphase separation, hitherto undescribed. These anaphases contain a different number of bivalents in the equatorial plane, but most univalents have already moved to the poles. Several such daughter-plates showed a high number of chromosome bodies, and the total number of these was greater than 36 (Figs. 40, 41). The interpretation is easy. Some pseudohomeotypic divisions contain a variable number of bivalents, and the chromatid repulsion of univalents is prior to bivalent separation. From a cyto-mechanical point of view this division type is very interesting. Division is delayed in comparison to the case in prophase type 1. Consequently chromosome pairing and chiasma formation can ensue. But at the same time prophase or prometaphase is retarded, giving the univalents time enough for a splitting of the centromere. In the subsequent meta- and anaphase ·chiasmata keep the bivalent-chromosomes together and their separation requires a greater force and a longer time than the chromatid separation. Therefore as far as univalent splitting is concerned anaphase separation is complete at a time when bivalents have not begun to disjoin. In some anaphases

the equator is still occupied by true bivalents and small splitting uni-
valents at the same time (Fig. 41).

As in *H. robustum*, chiasmata are exclusively terminal at meta-
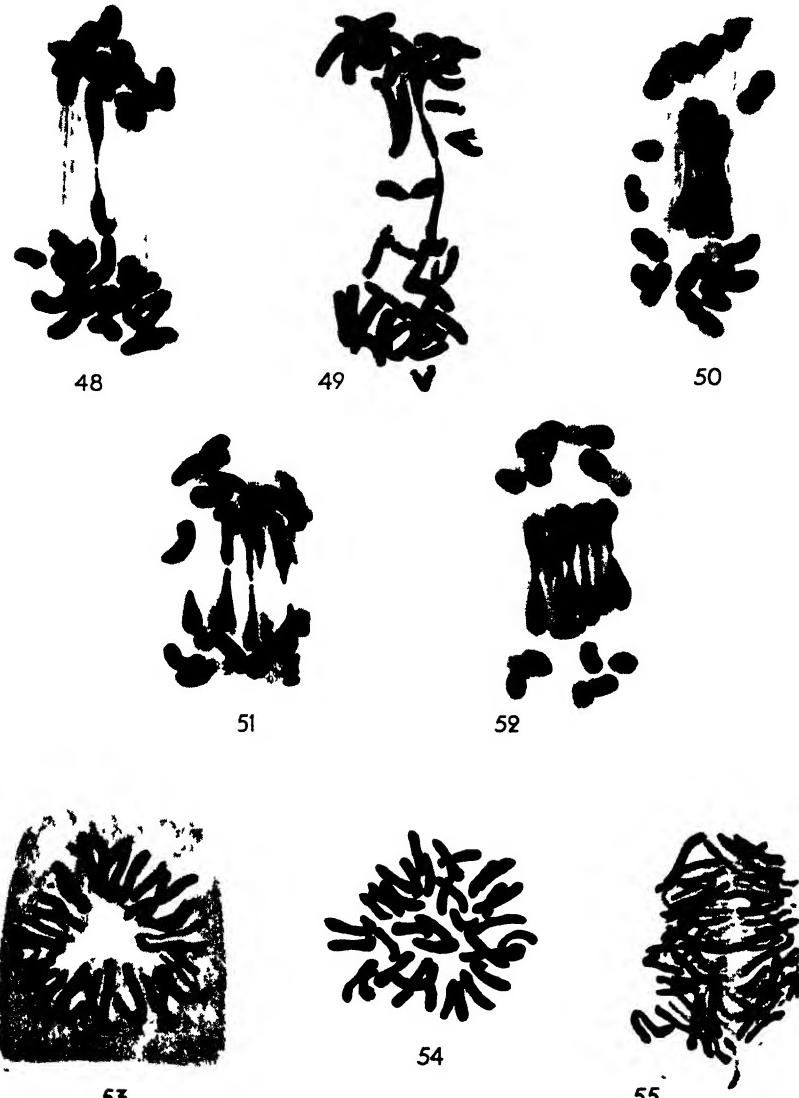
Figs. 48—52. Meta- and anaphases with different numbers of bivalents. Note the
variable chromosome contraction. — 53—55. Mitotic-like divisions after nuclear
contraction. — ✕2100.

phase. Inversion-bridges are common and frequently numerous (in
some cases 9—10 bridges have been observed, Fig. 43). This indicates

that even the pairing genomes are structurally different, and for that reason the designation employed (allogenomatic biotype) is justified. The terminal chiasmata and the occurrence of inversion-bridges argue in favour of a pachytene pairing by terminal sections or a terminalisation over non-homologous segments.

Telophase nuclei look different, whether they arise after semiheterotypic or pseudohomeotypic divisions, as pointed out by GENTSCHEFF in 1937. In the former case they are rounded off to all sides (Fig. 35), in the latter case they are half-moon-shaped, the linear surface turned inwards (Fig. 42). After bivalent formation telophase nuclei appear as in the second case. The cause of this different appearance is apparent. Semiheterotypic divisions contain scattered univalents; after the hypothetical stretching of the middle-part of the spindle some univalents still remain between the poles. Nuclear membranes are formed, including most of the chromosomes. Sometimes they give rise to restitution nuclei, but their edge will always be rounded off. In anaphases after high bivalent formation or in pseudohomeotypic divisions separation is regular and the chromosomes (or chromatids) reach the poles simultaneously. Therefore the aspect facing inwards will be sharp and linear.

Fragmentation in *H. amplexicaule* differs somewhat from the appearance in *H. robustum*. As mentioned above, it takes place in lateral flowers following prophase of type 2 with prolonged interphase stages (cf. *H. robustum* and *Leontodon hispidus*). However, it is never found in PMC:s containing bivalents but is exclusively associated with pseudohomeotypic divisions (Figs. 44—47). In the latter case univalents gather to divide already at first metaphase (which in this division type is also the second) after the centromere division has occurred. A certain stress on the chromosomes will arise at that moment. When the chemical changes of the chromosome structure begin (at late metaphase—anaphase), the breaking up of chromatids into smaller parts is first seen. Why this does not happen in the still later divisions containing bivalents is unknown to us so far. Probably some difference in the physiological condition of the preceding interphase is responsible for the change in stability. Fragmentation is more intense at the top of a pollen-sac, as in the case of *H. robustum,* suggesting some physiological influence by the PMC-location.

Fragmentation in *H. amplexicaule* is gradual, not sudden as in *H. robustum*. The first sign of a fragmentation is the occurrence of bodies having approximately half the univalent size. They orientate

in the spindle like splitting univalents at first anaphase. Chromosome bodies are also seen, equal in size to a quarter of a univalent, similarly arranged between the poles. The same thing is true of even smaller chromosome pieces. Since most of these parts of previous univalents lack centromeres, the regular and longitudinal orientation cannot be due to a centromere repulsion but to other forces acting in the spindle on the fragments. From the behaviour of X-ray fragments in plants we know that fairly small fragments, lying in the equator, frequently orientate in the length-axis (GENTCHEFF and GUSTAFSSON, 1939; cf. CARLSSON, 1938, for X-ray fragments in *Chortophaga*). The hypothesis might be put forward that chromosome fragments or parts — if lying in the middle, not on the outskirts of the spindle and showing tendencies to split — arrange regularly, irrespective of any possession of centromeres. The different behaviour of the middle and the peripheral parts of the spindle may be attributed to special properties associated with the stretching at anaphase (BELAR, 1929; DARLINGTON, 1932, 1937; GUSTAFSSON, 1935).

Another mode of fragmenting is shown in Figs. 44—46. Chromosomes are stretched out at anaphase, giving a bridge-like appearance. At a later stage the threads fall into numerous pieces almost simultaneously. By this fragmentation the chromosomes apparently break up into their ultimate chromomeres. After counting the number of fragments in a cell, it would be possible to form an opinion regarding the chromomere number. Owing to the very high number of small pieces we hesitated, however, in carrying out such a task.

In semiheterotypic divisions as well as in PMC:s with bivalent formation chromosome contraction varies widely (Figs. 49, 52). Metaphases with long and with short chromosomes exist. Usually the former type of division contains strongly condensed univalents, the latter type less contracted chromosomes. Long-shaped univalents seem to distribute more irregularly at anaphase. The highest chromosome contraction is seen in pseudohomeotypic divisions.

In some pollen-sacs transitional stages to the *pseudoillyricum* type are met (Figs. 53—55). Contraction stages of the nucleus occur at late prophase. These contraction stages are found also after semiheterotypic prophase (type 1). In those cases the univalents are still contracted at metaphase. Even metaphases of mitotic appearance — with long and slender chromosomes arranged regularly, as in mitosis — were found after slight nuclear contraction. These metaphases appear most

frequently after prophase type 2 or transitional stages between type 1 and 2.

Regarding prophase type 3, occurring in very old flowers, not much can be said, owing to the fact that later division stages were not found. Cells and nuclei have grown intensely already before mid-prophase. Chromosomes lie well scattered within the nuclear membrane and seem less contracted than in divisions of semiheterotypic character.

TAPETAL GROWTH AND DEVELOPMENT IN RELATION TO MEIOSIS.

In our study of tapetal development we included, for the sake of comparison, two sexual species, *H. speciosum* and *H. leiophanum*. Both are diploid (2n = 18), forming nine bivalents at meiosis. A third type, *H. caerulaceum*, is triploid (3x = 2n = 27), but shows an almost typical meiosis. Whether it is apomictic or sexual we cannot say. Pairing is very high at metaphase (11—12—13 bivalents and a few univalents). Apart from the occurrence of some micronuclei tetrad formation is regular. No careful examination of meiosis was made.

In these three biotypes the growth of tapetal cells was studied at different stages of meiosis and tetrad formation by measuring the length of tapetal cells in a pollen-sac. The number of PMC:s corresponding to one tapetal cell was calculated. At early meiotic stages the PMC:s lie close to each other. The division of the length of a pollen-sac by its number of PMC:s gives an average figure of the length of a PMC. In later stages when the PMC:s have grown intensely or the tetrad membranes are swollen, the same figure gives only the space that one PMC has at its disposal.

With regard to the designation of the different stages not much need be said. Early prophase denotes the stages up to pachytene (synizesis), mid-prophase is the stage when chromosomes begin to shorten and late prophase is approximately identical to diakinesis. Tables 2 and 3 give in a) figures of tapetal growth and in b) figures regarding the growth or space occupied by PMC:s. Each unit of the ocular micrometer corresponds to $1,3 \mu$. Several whole pollen-sacs or parts of them have been measured.

There is a remarkable growth period between early and mid-prophase at the time of pachytene pairing and chiasma formation in all three biotypes. In *H. speciosum* tapetal cells grow from 19,7 units at early prophase to 28,5 units at mid-prophase. In *H. leiophanum* the

TABLE 2.   The growth of tapetal cells (a) and PMC:s (b) at different stages of meiosis in Hieracium species with regular bivalent formation.

	Early pro-phase	Mid-phase	Late pro-phase	1 Meta-phase	Interkine-sis	Early tetrad	Ready tetrad	Late tetrad	Pollen
H. speciosum a) Tapetum	875 — 44 = 19,7 units	855 — 30 = 28,5 units	780 — 29,5 = 26,4 units	915 — 33,5 = 27,3 units	940 — 32,5 = 28,9 units	1300 — 45 = 28,9 units	1200 — 36 = 33,3 units	—	—
» b) PMC:s	825 — 102 = 8,1 units	855 — 78 = 11,0 units	780 — 74 = 10,5 units	900 — 82 = 11,0 units	880 — 82 = 10,7 units	1075 — 92 = 11,7 units	1200 — 80 = 15,0 units	—	—
H. leiophanum a) Tapetum	667 — 40,5 = 16,5 units	595 — 24 = 24,8 units	380 — 12 = 31,7 units	560 — 19,5 = 28,7 units	—	710 — 23 = 30,9 units	900 — 24 = 37,3 units	1500 — 33,5 = 44,8 units	900 — 19,5 = 46,2 units
» b) PMC:s	667 — 64 = 10,4 units	585 — 48 = 12,2 units	350 — 35 = 10,0 units	480 — 48 = 10,0 units	—	710 — 50 = 14,2 units	900 — 54 = 16,7 units	1500 — 89 = 16,9 units	—
H. caerulaceum a) Tapetum	1040 — 32,5 = 32,0 units	975 — 23 = 42,4 units	1145 — 29 = 39,5 units	605 — 15,5 = 39,0 units	—	—	—	1315 — 29 = 45,3 units	—
» b) PMC:s	710 — 50 = 14,2 units	975 — 70 = 13,9 units	635 — 39 = 16,3 units	490 — 28 = 17,5 units	—	—	—	1315 — 68 = 19,3 units	—

corresponding figures are 16,5 and 24,8 units (at late prophase the figure is 31,7). In *H. caerulaceum* they are 32,0 and 42,4 units. This growth period is marked by an intense mitotic activity in the tapetal cells. Tapetal cells, containing one nucleus, change this condition rapidly to two- and four-nuclear. In the sexual types there is a tendency of fusion between tapetal nuclei already at two-nuclear stage. But frequently fusion takes place in the four-nuclear stage. Tapetal cells with eight nuclei are also seen but more seldom. All these different stages of development appear simultaneously with the growth-period.

After the end of this growth-period the tapetum is remarkably constant, and only when tetrads are ready and their membranes dissolve do they show a second growth-activity. The figures for *H. speciosum* are 28,5 units at mid-prophase, 26,4 at late prophase, 27,3 at metaphase, 28,9 at interkinesis and 28,9 at early tetrad stage (before the final formation of four cells). The constancy is obvious. Later there is a slight increase to 33,3 units. In *H. leiophanum* the figures are 31,7 at late prophase, 28,7 at metaphase and 30,9 at early tetrad stage. At late tetrad stage and at pollen stage the tapetal length is 44,8 and 46,2 units respectively. In *H. caerulaceum* tapetal length is 42,4 units at mid-prophase, 39,5 at late prophase and 39,0 at metaphase, but at late tetrad stage 45,3 units.

Of considerable interest is the fact that the morphological doubling of chromosomes takes place at finished zygotene pairing at the time of chiasma formation, i. e. when tapetal cells (in *Hieracium*) have their greatest growth and division activity. The increase in volume of PMC-nuclei at meiosis has been referred to this stage (BEASLEY, 1938). At the beginning of prophase there is evidently a special growth and re-production impulsion spreading over the pollen-sac into the tapetum and PMC:s. In *Hieracium* only one row of PMC:s occurs, surrounded by the tapetum on all sides. An impulsion on the PMC:s via the tapetum is very likely.

The PMC:s themselves do not grow much, in contrast to their nuclei. From early prophase to interkinesis the length of the PMC:s in *H. speciosum* is 8,1, 11,0, 10,5, 11,0, 10,7, indicating a slight growth at pachytene. The figures for *H. leiophanum* are 10,4, 12,2, 10,0, 10,0. Here the increase is more obscure. Figures for *H. caerulaceum* are 14,2, 13,9, 16,3, 17,5, suggesting a growth process at mid-prophase. If a meiotic length increase exists, it is connected with a fairly early pro-phase stage. The growth values would probably be more marked if the increase in breadth was also taken into consideration.

The apomicts, examined here, behave quite differently. Semi-heterotypic division in both *H. robustum* and *amplexicaule* is carried out at a very early stage of tapetum development. At well advanced metaphase tapetal cells frequently contain one or two nuclei and fusion phenomena have rarely taken place. The growth curve is changed. At early prophase the tapetal cells are smaller in the tetraploid apomicts than in the diploid sexuals, in spite of the usual correspondence of polyploidy and cell size. In *H. robustum* tapetal cells cannot attain the size characteristic of *H. speciosum* or *leiophanum* (at interkinesis the values are 20,0 units as against 28,9 and $\pm$ 28,7). In *H. amplexicaule* tapetal cells do not grow up to 30 units until interkinesis and they never give the figures characteristic of the triploid examined, *H. caerulaceum* ($\pm$ 39,0). The mutual relation between meiosis-start and tapetum growth is entirely changed in the apomicts. Tapetal growth — when it occurs — takes place at a late stage of meiosis.

The figures for semiheterotypic division and PMC-growth are very noteworthy. The smallest size of sexual PMC:s at any stage examined is 8,1 for *H. speciosum*, 10,4 for *H. leiophanum* and 13,9 for *H. caerulaceum*. Early prophase cells of *H. robustum* are less than 6,5 units, those of *H. amplexicaule* less than 7,0 units, i. e. semiheterotypic meiosis begins at a stage of PMC-development when growth has not occurred or the dividing PMC:s have not acquired even the *diploid* size. Semi-heterotypic divisions start precociously in relation to tapetal development and PMC-growth.

Prophase type 2 in *H. amplexicaule* leads to pseudohomeotypic divisions and bivalent formation — and owing to inhibited chiasma formation to some semiheterotypic divisions. The tapetal cells in this prophase type (with synizesis-phenomena) pass through an activity period rather early and at first metaphase they have reached the four-fused stage. Their growth-curve is similarly expressed. At the end of early prophase they have an average length of 35,2 units, compared to 14,2 after prophase type 1 and 19,7, 16,5, 32,0 in *H. speciosum*, *leiophanum* and *caerulaceum*, i. e. they have grown considerably. The length of PMC:s is 12,9 as against 6,7 in prophase type 1. When bivalents are formed, the preceding tapetal growth and activity and even the PMC-growth are similar to those in sexual species.

At metaphase we find divisions which are pseudohomeotypic and sexual (with bivalent formation). Undoubtedly the pseudohomeotypic divisions show an intense prophase or interphase growth with regard to tapetum and PMC:s. Therefore the resulting univalents are given

TABLE 3. *The growth of tapetal cells (a) and PMC:s (b) in different division types of H. robustum and amplexicaule.*

	Inter-phase	Early pro-phase	Mid-pro-phase	Late pro-phase	Meta-phase	Interkine-sis	Restitu-tion nuclei	2nd divi-sion
H. robustum. Type 1. a) Tapetum......	—	135 — $9,5 = 14,2$ units	—	—	280 — $12,5 = 22,4$ units	150 — $7,5 = 20,0$ units	—	—
» b) PMC:s	—	135 — $21 = 6,4$ units	—	—	230 — $23 = 10,0$ units	150 — $14 = 10,7$ units	—	—
Type 2. a) Tapetum	1040 — $27 = 38,5$ units	1310 — $32 = 40,9$ units	—	990 — $24,5 = 40,4$ units	440 — $11 = 40,0$ units	—	—	—
» b) PMC:s	1040 — $89 = 11,7$ units	1300 — $107 = 12,1$ units	—	895 — $58 = 15,4$ units	440 — $26 = 16,9$ units	—	—	—
Type 3. a) Tapetum	—	—	—	—	640 — $13 = 49,2$ units	—	—	540 — $10,5 = 51,4$ units
» b) PMC:s	—	—	—	—	620 — $44 = 14,1$ units	—	—	400 — $25 = 16,0$ units

Prophase H. amplexicaule. type 1. a) Tapetum	860 — 60,5 = 14,2 units	1350 — 58,5 = 23,1 units	1460 — 62,5 = 23,3 units	730 — 31,5 = 23,2 units	1400 — 44,5 = 31,5 units
» b) PMC:s	860 — 128 = 6,7 units	1350 — 133 = 10,2 units	1460 — 144 = 10,1 units	730 — 71 = 10,3 units	1400 — 91 = 15,4 units
Prophase type 2. a) Tapetum	2200 — 62,5 = 35,2 units			740 — 23,5 = 31,5 units[1]	890 — 22 = 40,5 units
				165 — 4,5 = 36,7 units[2]	
» b) PMC:s	2200 — 170 = 12,9 units			740 — 61 = 12,1 units[1]	890 — 65 = 13,7 units
				165 — 11 = 15,0 units[2]	
Prophase type 3. a) Tapetum			340 — 10 = 34,0 units		
» b) PMC:s			340 — 24 = 14,2 units		

[1] Pseudohomeotypic divisions — [2] Bivalent formation

the time necessary for their ripening processes. That pseudohomeo-
typic divisions in *H. amplexicaule* imply transition-stages between semi-
heterotypic divisions and bivalent formation is shown by the fact that
they are irregular and incomplete after prophase type 1 but more
normal in character after prophase type 2. The remarkable division
type, still pseudohomeotypic but displaying a formation of bivalents,
proves this conclusion. Even the figures regarding tapetal and PMC-
growth indicate the same thing.

At metaphase we find the two above-mentioned division types (1
and 2). Tapetal cells in the case of pseudohomeotypic metaphases (1)
have reached a size of 31,5 units, i. e. they are slightly smaller than
the average sized cells at early prophase. Tapetal cells in pollen-sacs
with bivalent formation are, however, 36,7 units in length, i. e. these
cells are slightly larger than the average-sized cells at early prophase.
With regard to the PMC-growth cells giving pseudohomeotypic divisions
and bivalent formation the values are 12,1 and 15,0 units respectively,
as against 12,9 at early prophase. The following conclusion is therefore
arrived at: Pseudohomeotypic divisions are formed in those cells which
at early prophase certainly belonged to prophase type 2 but simult-
aneously were on the minus-side of the growth curve. Pollen-sacs,
giving this division type at metaphase, have not reached the same phase
of tapetum growth and development as in the case of bivalent formation.
Transitions between the two metaphase types exist.

In *H. robustum* loculi producing bivalents show an even more
advanced stage of tapetum development. Tapetal cells are 49,2 units in
length at metaphase, as against 36,7 in *H. amplexicaule* and 22,4 at
semiheterotypic metaphase. The length of the PMC:s is 14,1 units, i. e.
they have approximately the same size as corresponding cells in
H. amplexicaule. The mitotic activity of the tapetal cells is intense in
both species. At metaphase the formation of tapetal cells containing
four-fused or eight-nuclear tapetum is the rule. Bivalent formation
appears in sexual as well as in apomictic species exclusively when a
proper balance exists between tapetal divisions or tapetal and PMC-
growth and the onset of meiotic prophase. Disturbances in the course
of meiosis will result when any of the components in this balance
system is changed.

Another illustration of this result is given by the behaviour in the
event of double reproduction. Tapetal cells increase in length during
prophase and metaphase from 38,5 units to 40,8, 40,4 and 40,0 units, i. e.
with the exception of the first figure their size is fairly constant.

Apparently their most intense growth period takes place somewhere during the interphase. The values of PMC-length given in Table 3 are not the true ones. The length of a pollen-sac has been divided by the number of PMC:s occurring, but since several PMC:s grow just a little, some cells will be strongly compressed and their place taken by neighbouring PMC:s increasing intensely. The figures in Table 3 show that if all PMC:s were to grow equally, their size would nevertheless be greater at double reproduction than at bivalent formation (16,9 units as against 14,1). The length of tapetal cells is, however, very much greater in the latter case (49,2 units as against 40,0). At interphase and early prophase the figures recorded for PMC-growth (11,7 and 12,1 units) are considerably smaller than at late prophase and metaphase, but they are still higher than at interkinesis following semiheterotypic division.

In Table 1 the average size of ten actually dividing cells has been calculated. As already mentioned (p. 223), these figures show that a period of intense growth takes place at interphase. A second period of growth of nuclei and cells occurs at late prophase as a response to the doubled chromosome number. Tapetal activity is at an advanced stage already at early prophase, but the number of tapetal nuclei is not so high as in bivalent formation, nor is the growth of tapetal cells so well expressed. Owing to the incidental suppression of growth in some of the PMC:s, others are capable of an unusually great increase and so the double reproduction happens. Therefore, briefly summarized, PMC-growth is more advanced or intense in comparison to tapetal development and activity in the case of double reproduction than in the case of bivalent formation.

DISCUSSION.

This study has shown that in sexual species of *Hieracium* a special balance system exists with respect to the mutual relation of tapetum and PMC-development. The mitotic activity and growth of tapetal cells is especially pronounced at early meiotic prophase, the size of tapetal cells being constant from mid-prophase up to the moment when meiosis is completed and the pollen grains are formed. At this latter stage a second period of growth sets in, apparently associated with a swelling of cell-walls and cytoplasm without further mitotic activity.

In male organs of apomictic *Hieracium* types this balance system is more or less upset in different directions. A comparison between sexual and apomictic biotypes is carried out in this paper but a com-

parison is also made between the modes of tapetum development in different division types. Certain distinct features were found. Bivalents arise exclusively when tapetum growth, mitotic activity and PMC-growth are balanced to the prophase start in the same manner as in sexual species. ROSENBERG detected already in 1927 that bivalent formation is most complete in old PMC:s of lateral flowers. This finding was confirmed by GENTSCHEFF (1937). The eventual cause of the balance system, being properly adjusted chiefly in the lateral flowers, will be discussed later on.

The difference existing between meiosis in male and female organs of apomictic groups (GUSTAFSSON, 1938) can now be expressed in a manner not possible previously. *In the male organs* of *Hieracium* a tendency exists to increase the degree of precocity, upsetting the balance system of meiosis by the beginning of prophase before tapetal mitosis and PMC-growth take place. Semiheterotypic and pseudohomeotypic metaphases result, both types without pairing but with chromosome contraction. In the case of the highest precocity possible semiheterotypic divisions appear, giving rise to restitution nuclei rather frequently.

On the female side vacuolisation forces are generally active at the macrospore germination. An excellent account of the phenomenon in certain Rubiaceae-species was given by FAGERLIND in 1937. In several apomictic genera vacuolisation forces cause *aposporous* development from nucellar or chalazal cells. In the *Alchemilla* type of apospory potentially generative cells enter a meiotic prophase or show intense growth and vacuolisation at interphase stage and give mitosis. In female organs of *Hieracium*, finally, a tendency exists to decrease the precocity of meiosis, contrary to the case of increased precocity in male organs. Meiosis is omitted after an intense growth period and a long duration of the interphase. After *such a delay* semiheterotypic divisions are not formed, as far as we know, but pseudohomeotypic divisions frequently arise. After a certain threshold value of growth and vacuolisation division is entirely mitotic. An extreme case of such vacuolisation forces is met with in *Hieracium ramosum* (GENTSCHEFF, 1937), where diplosporous embryo-sacs are replaced by aposporous cells. The similarity of all these vacuolisation phenomena induced GUSTAFSSON to postulate a special hormone influence with regard to cell-elongation, similar to the effect of auxin. Evidently the different behaviour of male and female meiosis in *Hieracium* seems to depend on a different time-action of the prophase starting force in relation to growth and vacuolisation forces within the loculi and nucellus.

Another change of this balance system gives rise to the singular case of double reproduction in meiosis. In most loculi of *Hieracium robustum* prophase start and tapetum development show a better tuning than in semiheterotypic divisions but PMC-growth is too intensely expressed. Interphase cells and nuclei reach a size somewhat less than the metaphase volume in the case of bivalent formation. The present authors concluded that in *Spinacia* the double reproduction occurred at the transitional stage of interphase and prophase, being due to a disturbed ratio of cell and nucleus volume. That growth phenomena act as reproduction forces — cf. the nucleus plasm relation of HERTWIG — has now been assumed for the following cases:

In normal meiosis the morphological reproduction takes place at pachytene, at the stage of high nuclear growth. According to DARLINGTON and his followers pachytene implies the *actual* stage of meiotic reproduction. MARQUARDT (1937) showed that *physiological* reproduction at meiosis does not appear until the beginning of prophase (an accurate timing of the different prophase stages after X-raying seems difficult). SAX (1938) and MARSHAK (already in 1935) found a period of high meiotic susceptibility at early prophase (pachytene). Growth phenomena and delayed reproduction at meiosis are correlated, at least in a morphological sense.

• The cells of special tissues are capable of intense enlargement after auxin-treatment. As a consequence tetraploid numbers arise, chromosomes reproducing twice (LEVAN, 1939; GENTCHEFF and GUSTAFSSON, unpubl.). Growth phenomena precede the reproduction.

Double reproduction is frequent spontaneously in roots of *Spinacia*, where periblem cells contain tetraploid, octoploid and 16-ploid numbers. X-ray data indicate that the increase in number occurs predominantly at seed-germination as a response to high cell-growth.

An intense growth and vacuolisation changes the meiotic disposition in mitotic direction, as has been proved with respect to diplosporous and aposporous organisms (GUSTAFSSON, 1939). If growth and vacuolisation have been sufficiently high, interphase nuclei enter *mitosis* directly instead of meiosis displaying bivalent formation. Pachytene growth is omitted.

The chromosomes of those PMC:s in *Hieracium robustum*, which at interphase had the opportunity of cell-growth above a certain threshold value, reproduce twice, giving a metaphase with 36 pairs of univalents or 72 scattered chromosomes.

Of special interest is the fact that in spite of the double reproduction

occurring the PMC:s give divisions, meiotic in character. At metaphase chromosomes are usually contracted, almost round in shape, and lie scattered over the spindle, division being potentially reductional. Accordingly the meiotic force acting on these double-reproducing PMC:s is so strong that one extra reproduction, probably occurring at the growth-period at interphase, cannot cancel or balance it. If this interpretation is correct, meiosis can happen even if reproductions occur at interphase. A special balance system of meiotic and mitotic forces exists. An increase in the strength of the growth and reproduction force will change the character of division. If the meiotic force increases at the same time in effect, it will eventually counteract the mitotic disposition again. Whatever the explanation, a doubling of the chromosome number takes place in the form of internal reproduction, meiosis still being prevalent.

In the *Hieracium*-apomicts examined bivalent formation is restricted to the lateral flowers and cannot arise — even as a rarity — in the semiheterotypic or double-reproduction divisions in the median flowers. This shows that either the process of chromosome pairing or that of chiasma formation is entirely out of the question. If this depends on a shortened early prophase or on a changed time for chromosome splitting cannot be determined. One suggestion will be made. In the case of bivalent formation prophase is of the sexual type. Cell and nucleus increase in size at zygotene-pachytene. Presumably the duration of prophase is also increased. These two part-phenomena might facilitate the mechanical process of gene- and chromomere-attraction in pairs but also render the pairing firm and complete at the time when chromosome splitting occurs, essential for chiasma formation. This prophase growth might also facilitate the complete stretching out of chromonemata, after which the pairing can be accomplished. In the case of a rapid transitional stage of interphase and metaphase this uncoiling process may be omitted. Why bivalent formation in all species studied, whether sexual or apomictic, is dependent on a special tuning of the tapetum to the PMC:s, is impossible to explain fully.

Pronounced fragmentation processes are found in both apomicts. In *H. robustum* they are correlated to the formation of bivalents, in *H. amplexicaule* to pseudohomeotypic divisions. Fragmentation assaults the whole chromosome complement, bivalents as well as univalents. At incipient and advanced anaphase many cells are filled with small chromatin pieces, bivalents, trivalents or univalents suddenly or gradually breaking down. Previous studies on corn (BEADLE, 1932) have demonstrated that a recessive gene »st» changes the chromosomes

behaviour. The chromoplasm becomes sticky at meiosis as in the primary effect after high X-raying. At meiotic meta- or anaphase fragmentation is intense. In mitosis disturbances are not so frequent. Apparently a physiological difference between the meiotic and mitotic stages exist. In the two *Hieracium*-apomicts studied fragmentation is connected with a prolongation of the previous interphase as is the case in *Leontodon hispidus* (BERGMAN, 1935). Since a gradual fragmentation takes place at the onset of anaphase in *H. amplexicaule* and finally hundreds of chromomere-like bodies result, this might indicate — if the parallelism with salivary chromosomes is allowed — that the inert material between the chromomeres (nucleic acid-protein discs?; CASPERSSON, 1936) has not reproduced properly or is weakened. Nevertheless the chromonemata are kept intact by the substances, formed at prophase, providing a fairly stabile chromatin cover. At anaphase, when repulsion forces act on the chromosomes (cf. MATHER, 1934) and these revert to the interphase condition, the stability of the chromoplasm will disappear. At any rate physiological differences exist within the head. Interphase stages differ in duration and metabolism and consequently give rise to disturbances in the reproduction mechanism or the consistency of the chromonemata.

The authors dare not regard the balance system of meiosis as described in this paper as valid for phanerogams in general but should like to emphasize the view that future investigations of meiosis in *Hieracium* and other apomictic genera cannot refrain from taking into consideration the physiological influence on meiosis from or via the tapetum cells. That is noteworthy especially of organisms where PMC:s form one cell-row only, covered with tapetum cells on all sides. The correctness of this view is supported by STEBBINS and JENKINS's discovery in a *Crepis*-apomict (1939), where the tapetal cells form large undividing bladders and PMC:s degenerate already at early prophase. Tapetal cells are mitotically inactive, consequently PMC:s cannot develop — or vice versa. — Meiosis in hybrids, in inbred strains of cross-fertilizers, in homozygotes of genes causing asynapsis or sticky chromosomes is presumably partly associated with physiological disturbances, as outlined above. — Recently, OEHLKERS (1937) showed that another property of the plant organism, i. e. the constitution of chloroplasts or the amount of chlorophyll, influences the course of meiosis. Is this influence dependent on or parallel to a different development or function of the tapetum, and is this influence different in male and female organs?

SUMMARY.

1. Five *Hieracium* types, growing in the Botanical Garden of Lund University, have been examined with regard to chromosome number, meiosis, tapetum development and PMC-growth.

2. *H. speciosum* HORNEM. and *leiophanum* DT. are sexual diploids (2n = 18) with regular bivalent formation (9_{II}), *H. caerulaceum* ARV. is triploid, but shows a very high pairing at meiosis ($11—13_{II}$), *H. robustum* MARTR. and *amplexicaule* L. are tetraploid (2n = 36) and display a series of degeneration phenomena.

3. *H. robustum* gives three metaphase types at meiosis: 1) semi-heterotypic division, starting precociously with respect to tapetum activity and PMC-growth, 2) double reproduction, giving rise to an increase in the chromosome number at interphase (2n = 72), 3) bivalent formation, the maximum number of bivalents being approximately nine to eleven.

4. *H. amplexicaule* shows two types of prophase behaviour, corresponding to different metaphase appearance. The first prophase type is mitotic-like and gives rise to semiheterotypic divisions, the second type is sexual with a sensitive stage at early prophase (synizesis) and gives rise to regular pseudohomeotypic divisions and bivalent formation ($0—11_{II}$).

5. In the diploid and triploid types examined a special balance system of meiosis exists. Tapetal cells show a lively mitotic activity and an intense growth period at pachytene stage. PMC-growth occurs at the same time but is not so well expressed. After early prophase mitosis and growth of the tapetal cells are concluded, and a second period of growth does not appear until late tetrad stage but without any mitotic activity. At early prophase tapetum contains usually four single or fused and even eight nuclei.

6. In division type 1 of *H. robustum* division starts when tapetal cells are one-nuclear and very small, and PMC:s have not acquired the size characteristic even of the *diploid* species. Prophase is remarkably precocious.

7. In loculi displaying double reproduction tapetal cells and PMC:s show a better balance than in division type 1, but PMC:s nevertheless grow too intensely. In response to the changed proportion of cell and nucleus volume the chromosomes are forced to reproduce twice, as in *Spinacia*. The origin of 36 pairs of chromosomes is the result, each chromosome possessing one centromere and two chromatids. At meta-

phase the chromosomes lie in pairs (36_{II}) or have a scattered position (72_I). Division is meiotic in character in spite of the internal reproduction, metaphase being typically semiheterotypic.

8. Bivalent formation occurs exclusively when a proper balance exists between tapetal growth and activity, PMC-growth, prophase onset and chromosome reproduction. The balance system is identical in sexual diploids and tetraploid apomicts.

9. In *H. amplexicaule* prophase type 1 occurs when the tapetal cells are at the one- or two-nuclear stage and PMC:s still very small. Semiheterotypic metaphases result. Infrequently transitions to pseudohomeotypic divisions are found.

10. Prophase type 2 occurs at an advanced tapetum stage (four- and eight-nuclear stage). PMC:s are larger than in type 1.

11. Pseudohomeotypic divisions arise in those cells of prophase type 2 which at early prophase belonged to the minus side of the growth curve. They occur abundantly and are generally regular in character.

12. Bivalent formation appears after prophase type 2 in those cells which at early prophase were on the plus side of the growth curve. As in the sexual types and in *H. robustum*, a proper balance of tapetum and PMC:s must exist before bivalents will be able to form.

13. A division type, previously undescribed, has been observed, combining features from the pseudohomeotypic division with bivalent formation. Univalents and bivalents gather at the equator and divide. Usually the splitting of the univalents is accomplished when bivalents remain in the equatorial plane.

14. Bivalents have generally one terminal chiasma. Inversion-bridges are common at first and second division. In *H. amplexicaule* they are often numerous, nine or ten in number.

15. Fragmentation phenomena exist in both *H. robustum* and *amplexicaule*, in the former apomict connected with bivalent formation, in the second type connected with pseudohomeotypic divisions. They always occur after a prolonged interphase stage. Certain distinct features are described. Divisions are more regular close to the ovule than farther towards the top of the loculi.

16. In *H. robustum* degeneration of pollen-sacs sometimes starts very early. In such cases the tapetum also degenerates, the chromatin being discoloured and large vacuole-like formations being present. An obvious parallelism exists.

17. The significance of the balance system found in *Hieracium* is discussed.

18. The meiotic differences generally occurring in male and female organs of the same apomictic individual is expressed in the following manner: In *the male organs* a tendency exists to increase the degree of precocity, upsetting the balance system of meiosis by the beginning of prophase before tapetal mitosis and PMC-growth take place. On *the female side* growth and vacuolisation phenomena cause a lower degree of precocity, divisions generally not occurring until a great prolongation of interphase and an intense growth of the EMC and its nucleus have taken place. Evidently the different behaviour of male and female meiosis in *Hieracium* seems to depend on a different time-action of the prophase starting force in relation to growth and vacuolisation forces within the loculi and nucellus.

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CONTENTS.

THE CULTIVATION OF PLANT SPECIES FROM SEED TO FLOWER AND SEED IN DIFFERENT AGAR SOLUTIONS

BY G. GENTCHEFF AND Å. GUSTAFSSON

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IN the late spring of 1939 the present authors started a series of injection experiments with different hormones in order to study their effect on meiosis. Injections were also performed on the recessives and heterozygotes of the gene »sticky» in corn, producing a profound fragmentation of chromosomes at meiosis and changing the properties of the chromoplasm similar to that occurring after intense X-irradiation (the primary effect). Simultaneously we began to examine the physiological and hormonal causes of the double chromosome reproduction in periblem cells of *Spinacia* (GENTCHEFF and GUSTAFSSON, 1939). As in the case of *Allium* (LEVAN, 1939), auxin influences the growth of cells in certain tissues outside the ordinary root-meristems and as a consequence the chromosome number is doubled. The effect of auxins on the ordinary root-meristems was nil, not even on the periblem cells, where growth and increase in chromosome number take place spontaneously.

Another series of injection experiments dealt with the possibility of changing the type of embryo-sac development. As is well-known, the most common type of embryo-sacs originates from the development of one macrospore after the degeneration of the other three macrospores. However, some aberrant types are known in which the embryo-sacs develop from two or four macrospores. In genera having these latter types frequently intense growth and vacuolisation phenomena take place very early. Therefore we decided to study the influence of auxin, male and female hormones on the female development.

Owing to other research work we have not been able to study the injection material so far, but since another method was detected, by means of which plant species can be cultivated in hormonal media during the whole life-cycle, we regard this latter method as superior. The artificial cultivation implies a *normal response* of the plant under certain controlled environmental conditions even if the hormone or salt concentrations are sometimes detrimental to the organism.

For the sake of an experimental attack on the cultivation of germinating and excised roots we started on June 14th some experiments with sterilized *Pisum* and *Spinacia* seeds, cultivated in agar solutions, according to BONNER and ADDICOTT's formula (1937). To these agar solutions different auxins, colchicine, aneurin, folliculin and testosteron were added. We are indebted to the Rockefeller Foundation, Paris, for the supply of the three last-mentioned hormones.

BONNER and ADDICOTT's prescription, worked out mainly for excised roots, is valuable also for the following kind of work and is as follows (slightly changed):

$Ca(NO_3)_2$ ..... . 142 mg. per liter
$MgSO_4.7H_2O$ .... 36 » » »
KNO_3 81   ›   ›
KCl .. .. 65 ›   »
KH_2PO_4 .. .. 12 »
$Fe_2(SO_4)_3$ . ... 2 ›
Saccharose 4 %
Agar 0,5—1,0 %.

In the first experiment, mentioned above, no aneurin was used. Later we added aneurin to produce a 0,0005 % solution.

The sterilization of the seeds was first made in accordance with BONNER and ADDICOTT's suggestions. Later we found that 5 minutes' treatment in alcohol (96 %) and 15 minutes' treatment in sublimate (0,1 %) gave the best results for *Pisum*. For *Spinacia* the correspond-

Fig 1 *Pisum* plants grown under light conditions *a*) Without hormones, *b*) with folliculin, *c*) with testosteron Flower formation in all three tubes Note the size differences — $1/6$

ing figures are 2—3 minutes in alcohol and 10 minutes in sublimate. As *Spinacia* seeds are difficult to sterilize, we took off the seed coat. Parallel experiments were performed in darkness and in light. The temperature was 21° C.

In the experiment started first with the *Pisum* variety »American Wonder» flowers were formed under dark conditions on July 10th, i. e. after 26 days. These and some other experiments were carried out at the Botanical Laboratory of Lund University. We are indebted to

Professor Dr H. KYLIN for working place. Two weeks after the commencement of our investigation, but independently of us, BORGSTRÖM began at the same institute a *physiological* attack on the problems of flower formation in agar media, a report of which has already been published (1939).

After the preliminary work on the cultivation methods more extensive studies were started at the Institute of Genetics, Svalof, in the beginning of September. Several data have already accumulated. Pollen examinations and fixations have been made   Studies on the artificial

Fig 2   The same under higher magnification   Fruit formation in *b*) — ⁴/₆

cultivation of non-viable chlorophyll mutations in *Pisum* have begun, the seeds kindly supplied by Dr J. RASMUSSON, Landskrona.

Of the results obtained we shall only mention here that different varieties of *Pisum* behave extremely differently   The variety »Gulärt 36/23» did not form buds until 75 days after the beginning of the experiment (light). In the variety »Gråärt Å 33/205» flowers appeared in light conditions after 40 days and in darkness after 45 days. The variety »Extra Rapid», finally, formed buds in dark and light conditions after 21 days. »Gråärt» is a fairly early variety, but not so early as »Extra Rapid» or »American Wonder». Other late varieties have also been tested, which do not produce flowers easily either in dark or in

light conditions. Flowers are formed in control experiments without hormones, as well as in media containing auxins, folliculin and testosteron, but so far not in colchicine media. Fruits and seeds have been produced in folliculin and control media.

Flowers have also been produced in dark experiments with *Spinacia oleracea* (the variety »Hertha», kindly supplied by Dr. O. GELIN, Landskrona). Since *Spinacia* is dioecious, the cytological effect of folliculin on tetrad formation and that of testosteron on female meiosis and embryo-sac development will be especially interesting. An examination of the morphological effect of the reverse sex-hormones has not yet been made, because all flowerbuds obtained have been fixed at an early stage. The microscopical studies will reveal the eventual changes in the sex-expression.

Fig. 3. Flower formation under dark conditions without any hormones. Note the changed morphology of the flower. Anthers are free from each other — 2,5/1

The artifical cultivation of barley, wheat and *Allium* species was unsuccessful, due to the more difficult sterilization of seeds and onions. Since early varieties occur in barley, easily producing seeds under greenhouse conditions after three months, a satisfactory sterilization will probably yield positive results.

The cytological and genetical significance of this cultivation method is obvious. Recent results obtained by OEHLKERS and his co-workers on the effect of environmental changes on the chiasma formation and the process of terminalisation have shown that various exterior agencies influence the course of meiosis (cf. OEHLKERS, 1937). Especially noteworthy in this connection are the results in changed water balance systems and in different plastid environment. We have found that seeds of different *Pisum* varieties can germinate in sugar concentrations from 0—18 % and are capable of growth. In this way a variable osmotic pressure can be produced in the environment of the plants, secondarily influencing the osmotic pressure and the water balance inside the organism. In the female organs of several apomictic genera changes in the meiotic disposition

are connected with growth and vacuolisation phenomena. In male organs of *Hieracium* apomicts bivalent formation is connected with a special balance system of tapetum activity, PMC-growth and prophase start (GENTCHEFF and GUSTAFSSON, 1940). The experimental change of this balance system in other plants is our ultimate purpose.

OEHLKERS and co-workers have shown that reciprocal *Oenothera* crosses, producing identical genotypes, nevertheless give differences with regard to the constitution of plastids and the mutual amount of carotin + xanthophyll and chlorophyll substances. These differences are responsible for some changes in the course of meiosis (low chlorophyll content corresponds to a decrease in the number of chiasmata or an
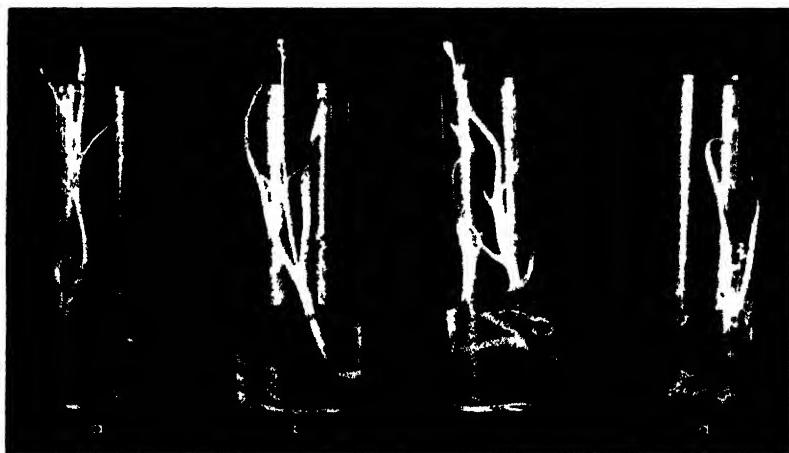
Fig. 4. Tubes with *Spinacia* cultures in dark conditions. In *a*) colchicine, in *b*) testosteron, in *c*) and *d*) different auxin concentrations. — $^1/_1$.

increase in the terminalisation, »Bindungsausfall»). With the method described above it will be a simple matter to find out whether this change in meiosis is dependent on the actual structure of the plastidogen and the plasm itself or only on the present amount of chlorophyll. The cultivation in darkness and in light simultaneously will solve the problems with regard to the genotype in question. In *Pisum* a number of different chlorophyll mutations have been described (RASMUSSON, 1938), consisting of non-viable as well as viable types. The parallel cultivation of the recessives, compared with the heterozygotes and the normal homozygotes, will reveal the significance of the genotypical changes of the plastid structure for the course of meiosis, assuming, of course, that the mutations tested have arisen from early varieties possible to cultivate.

The parallel behaviour of genetical earliness and the possibility to produce flowers on agar will, it seems to us, be of importance in plant-breeding work. In three to four weeks it may be found out whether a cross is transgressive with regard to earliness or not. Since only a small number of seeds is required for the cultivation and this can be performed with agar lacking any hormones, it implies an increase of the plant-breeding facilities. As mentioned above, the present authors have started a series of careful experiments with ten different *Pisum* varieties.

In several dioecious genera, such as *Melandrium* and *Spinacia*, hermaphrodites are known. In *Melandrium album* and *rubrum* and in *Spinacia* (cf. ÅKERLUND, 1928 and HAGA, 1935) the male sex is heterogametic. In *Melandrium* an XY-pair exists, but no difference between the two sex-chromosomes has been found so far in *Spinacia*. Most of the hermaphrodites in *Melandrium* have the XY-constitution, occasionally the female sex is changed (ÅKERLUND). In *Spinacia* the case is quite different. ROSA (1925) found a frequency of 0,2 % intersexes among 5198 plants. According to him, nutrition conditions, light, space, mutilation and so on, do not influence the number of intersexes. HAGA (1938) found, however, a much higher frequency of hermaphrodites, varying from 3,7 to 18,8 %. In all, 105 plants in an offspring of 1307 individuals were hermaphrodites. It seems obvious to us that special *internal* conditions change the female appearance, the intersexes being genetically homogametic and female. The finding that *Spinacia* may be cultivated as easily as *Pisum* (the buds in Figs. 4 b—d were produced after 20 days; cultivation in complete darkness), makes it possible to approach the physiological causes of secondary hermaphrodism in plants.

Physiological problems, which are of interest to us, concern the possibility of cultivating chlorophyll mutations of *Hordeum* and *Pisum*. Viable *chlorina-* or *viridis*-types will probably form flowers quite readily, but *albina-* and *xantha*-types are presumably more difficult to cultivate. Some preliminary experiments have failed. Since *albina*-mutations (in *Hordeum*) change the situation of their stomata and decrease the transpiration (GUSTAFSSON, unpubl.), new attempts will be made in order to solve some problems regarding xerophyte and saprophyte structure.

Svalöf, November 28th, 1939.

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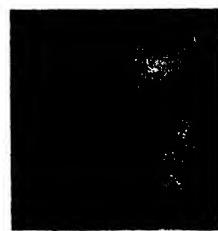
TWO EXTREME X-RAY MUTATIONS OF MORPHOLOGICAL INTEREST

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AMONG a series of X-ray mutations in barley, obtained in the experiments of recent years, the first-mentioned author found two types, which are of interest from a morphological and phylogenetic point of view. According to the latter author, working on the taxonomy and phylogeny of *Hordeum*, these types are not previously met with in the literature.

Both mutations have been obtained from a pure line, Golden barley (»Gullkorn»), isolated at Svalöf already before 1900. Golden barley is a typical *Hordeum distichum nutans* type, of great importance to the Scandinavian barley breeding. The first mutation (the two-flower type) arose in the X_2-generation in 1937 from seeds irradiated very intensely in the spring of 1936. The X_1-generation showed a germination capacity of 31 %. The second mutation (with lemma-like glumes) arose in the offspring of a specially treated seed-series. 645 seeds were desiccated above conc. H_2SO_4 and then X-rayed. In the roots of some germinating seeds the frequency of chromosome disturbances was examined and, in fact, most cells in division contained fragments and two-centromere formations. The germination capacity was 22 % and only 7,7 % fertile plants were formed. The sterility in the X_1-generation was high, and the mother-plant of this mutation, which appeared in the X_2-generation, showed 66 % sterility.

The two-flower mutation. — Within each ordinary lemma two flowers are formed. This type is completely sterile and segregates as a recessive. The heterozygote cannot be distinguished from the dominant Golden barley-type. The segregation is in the ratio 3 : 1. In 1938 the actual segregation was 413 : 124 (D/m approximately equal to 1). The cause of the sterility is unknown. — The long-awned primary lemma is normal. The barbs of this lemma are less numerous and less pronounced than in Golden barley (3—4 per nerve). The spikelets of the middle-row contain two flowers, the lemmas of which have 4—7 barbs each, thus in closer agreement with the mother line. The awns
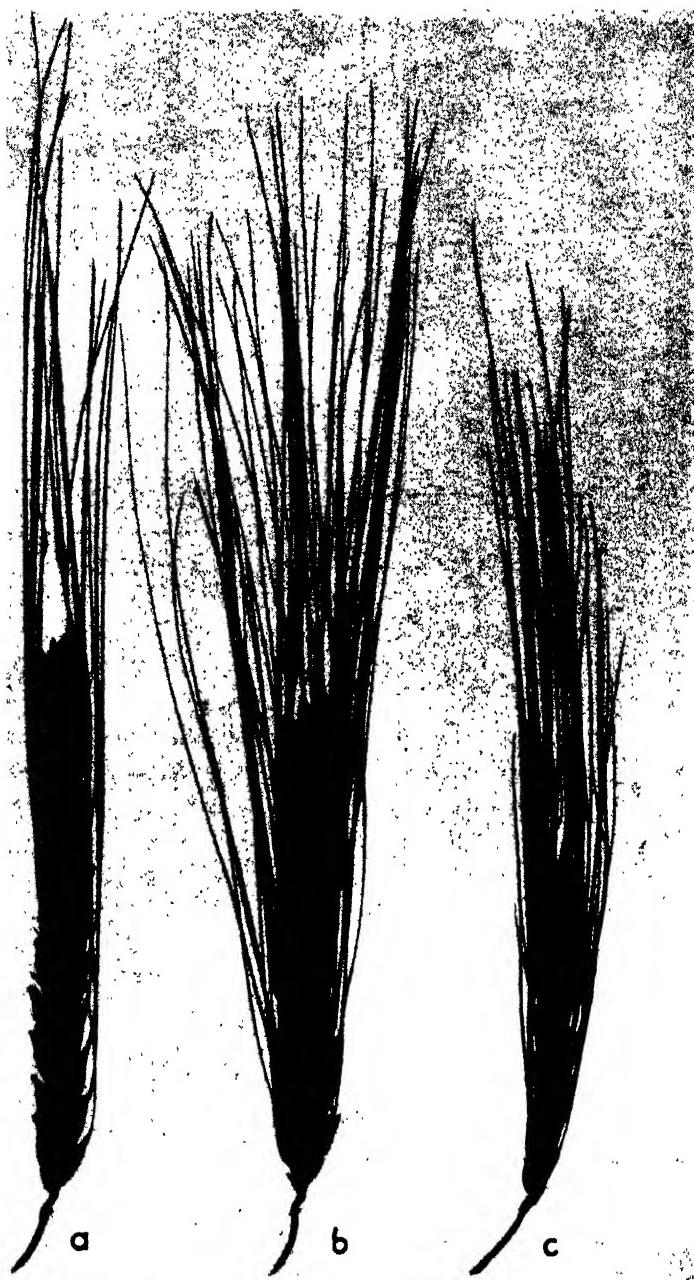
Fig. 1 a: Ear of Golden barley, b: of a mutation with lemma-like glumes, and c: of a mutation with two flowers within the primary (ordinary) lemma. — $^1/_1$.
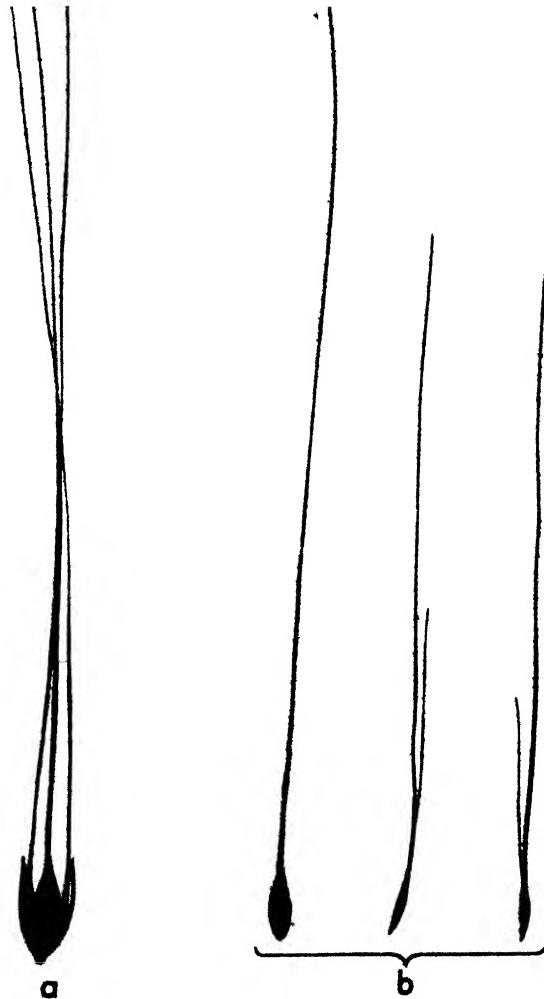
Fig. 2 a: Flower with lemma-like glumes and three long awns   b: To the left: the primary or **ordinary** lemma, to the right: the two individual flowers within such a primary **lemma**, each flower with one long and one short awn — $^1/_1$.

of the secondary lemmas **are** fairly fine, about $^3/_4$ of the length of the awns on the primary lemmas.   The paleæ have short awns about 2 cm.

in length. The barbs of the awns from all lemmas and paleæ agree in appearance with those in Golden barley. The rachillas are normal with long hairs.

A mutation with lemma-like glumes. — Due to its complete fertility and constancy this type is of direct breeding interest. Seeds are unusually large. Outer glumes look like lemmas, each of them having an awn of the same length as that of the lemma. (In Golden barley the glumes are linear with short awns.) The size of glumes and lemmas is identical. In contrast to the case in Golden barley the outer glumes are entirely without hairs. Barbs are missing. Barbs of the lemmas, however, are as numerous and pronounced as in Golden barley. All three awns have barbs. The lemmas in the side-rows are either blunt or pointed with or without short awns of about 1 $^1/_2$ cm. in length.

The anatomical structure and ontogenic origin of the two flowers within the primary lemmas in the first-mentioned mutation is so far unknown to us, and we have seen no taxonomical descriptions in the literature corresponding to this type. SCHIEMANN (1921) described the anomalous occurrence of two-flowered spikelets in the progeny of a special barley-cross. In a sample of *Hordeum vulgare* L. var. *pallidum* SÉR., VAVILOV found an unknown variety, differing from the normal in having a third outer glume at the base of the ear and small leaflets at the base of middle grains in the ear (VAVILOV and BUKINICH, 1929). This variety, named *afghanicum* VAV., is however only superficially similar to the one described here.

No cultivated variety or species of barley hitherto known from Europe or America shows the characters of the second mutation with regard to the lemma-like outer glumes. BOSE (1931) described a spontaneous variety of Pusa barley with broad outer glumes possessing awns and in 1937 he wrote about the heredity of this character. Whether the mutation with lemma-like glumes is identical to the variety studied by BOSE with regard to the properties of the glumes is difficult to tell. This mutation seems to indicate that the differentiation of outer glumes and lemmas has not become completely stabile. The similarity of this mutation to a variety of *Hordeum spontaneum* C. KOCH, named *ischnatherum* COSSON, is obvious, if the properties of the lemmas in the side-rows are taken into account. In his paper of 1908, KÖRNICKE has directed the attention to this feature of var. *ischnatherum*. The lemmas of the side-flowers in *H. spontaneum* are blunt as in cultivated *H. distichum* L. In the variety mentioned they were

either pointed or short and thin-awned. SCHWEINFURTH (1908) was of the same opinion. SCHULZ (1912) does not agree with KÖRNICKE and regards *ischnatherum* as a step forward from *H. spontaneum* to the six-rowed barleys. According to him, *H. spontaneum* var. *ischnatherum* has not only pointed or short-awned lemmas, but these can even be blunt. The X-ray mutation described above shows that pointed and short-awned types may arise from normally blunt varieties. That might be the cause of the origin of such variations in nature.

These mutations are of interest from a plant-breeding point of view. They show that it is possible to produce mutations outside the normal variability of cultivated and high-yielding plants. Especially the second mutation, which is completely fertile and viable, may be mentioned. Seeds are larger than in Golden barley, presumably due to an increased assimilation around the flowers. In a paper to be published shortly, one of the authors will describe a series of viable mutations produced after special treatments by X-raying. They fall, however, within the normal variation sphere of the cultivated barley. Planned viability and production tests will show whether these viable mutations are inferior, equal or, in some cases, even superior to the mother-line.

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THE EFFECT OF ACENAPHTHENE AND COLCHICINE ON MITOSIS OF ALLIUM AND COLCHICUM

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THE effect of colchicine on the chromosome mechanism is of a distinctly different nature from the pathological phenomena caused by most other poisonous substances. The most characteristic feature in the action of colchicine is perhaps its extremely keen selectivity. While most other poisons bring about mitotic disturbances in general, colchicine affects only the spindle apparatus. Other vital processes of the chromosomes seem to continue normally, even during colchicine treatments of considerable length. In view of this fact it seems necessary to assume that the colchicine molecule has a specific ability to act directly upon the molecules which build up the spindle organs, the centrosomes and centromeres, while other physical and chemical agents cause more or less complex disturbances of mitosis. In order to stress this point I introduced the term c-mitosis to denote the abnormality of mitosis caused by colchicine.

It soon turned out that other substances as well as colchicine were able to cause c-mitosis. Thus SCHMUCK (1938), KOSTOFF (1938 a) and NAVASHIN (1938) reported that they had found another substance, acenaphthene, which had a similar effect to that of colchicine. And later on SCHMUCK and KOSTOFF (1939) and SIMONET and collaborators (for instance, SIMONET and GUINOCHET, 1939) directed their attention to several cyclic halogene derivatives with similar action. NEBEL (1938), however, failed to get any effect of acenaphthene in his material, staminal hairs of *Tradescantia*. And BLAKESLEE (1939) states that acenaphthene »seems relatively ineffective in inducing 4n plants in the species with which we have tested it» (p. 163).

If the effect of colchicine is due to a very specialized reaction, a temporary poisoning or inactivation of the spindle organs, it is *a priori* surprising that these relatively simple benzole and naphthalene derivatives are able to produce the same effect as the very complicated colchicine molecule. It therefore seems appropriate to make a critical comparison of the action of colchicine and these other substances on a

cytologically suitable material. The present paper deals with the effect of acenaphthene on the root chromosomes of the same *Allium fistulosum* clone, the reaction of which to colchicine was studied earlier (LEVAN, 1938). Its normal cytological conditions have been controlled for several years. In order to elucidate the problem also from another angle, the effect of colchicine and acenaphthene on *Colchicum* is described afterwards. *Colchicum* should for natural reasons be immune to colchicine. And BLAKESLEE (l. c.) met with a negative result of the macroscopical c-tumour reaction in *Colchicum*: »It is like the snake which is immune to its own venom» (p. 163). If *Colchicum*, in spite of its immunity to colchicine, is affected by acenaphthene the possibility must be considered as to whether two different processes necessary for normal spindle function are present, each of which may be affected separately, one by colchicine and the other by acenaphthene.

Owing to the insolubility of acenaphthene in water, the treatments of root tips with acenaphthene must be performed in a somewhat different way from the colchicine treatments. The smaller *Allium* bulbs were treated with acenaphthene by wrapping the roots in moist filter-paper and putting them in petri dishes, in which small amounts (about 1 g) of acenaphthene were placed. The larger *Colchicum* bulbs were placed in an acenaphthene atmosphere under a glass-bell. It was found that a positive effect could be obtained by placing the acenaphthene crystals on small watch-glasses at a short distance from the roots. Thus the vapour of the substance is effective. In order to obtain the maximal effect quickly, the best method, however, is to dust the roots directly with plenty of acenaphthene and then wrap them in moist filter-paper. This very strong treatment was not lethal as a rule, either in *Allium* or in *Colchicum*. Since the course of the changes in the mitoses were more regular following such maximal dosages, most of the experiments described below refer to such treatments.

My thanks are due to Dr. O. HAGERUP, Copenhagen, for providing me with *Colchicum* material, and to Miss M. PALM for valuable technical assistance.

I. ACENAPHTHENE EXPERIMENTS WITH ALLIUM.

A clear difference in action between colchicine and acenaphthene is observed in the very first appearance of the disturbances. As soon as the concentration of the colchicine solution tested is increased above the threshold value, the effect sets in with remarkable suddenness and com-

pleteness. In fact, the effect is maximal already a few minutes after the beginning of the treatment, and normal mitoses are entirely absent from the tissue. Acenaphthene acts decidedly more slowly, its initial period from the first visible action until full effect lasts for several days. Even after a treatment of 5 to 7 days plenty of normal anaphases may be found in the tissue and only after the longest treatments in my experiments was the tissue completely devoid of normal mitoses. On account of this extension of the initial period, we have a splendid opportunity here of studying the induced changes step by step in a manner impossible after colchicine treatments.

After a treatment of 4 to 24 hours most mitoses take place normally. A number of quite normal anaphases are seen exceedingly well in the longitudinal sections. Still the first signs of abnormalities are already present. The equatorial plates have not always developed quite regularly. One or two chromosomes may be located outside the plate, or the whole plate may be changed into a half-spherical shape, indicating that one of the centrosomes has been put out of function.

During the next period, a treatment of 1 to 4 days, more conspicuous changes of the spindle appear. These changes almost always begin in the exterior part of the spindle, in the centrosomes. Thus the congression of the chromosomes into the equatorial plate becomes more and more deficient. And when no trace of an equatorial plate can be detected any longer and the chromosomes, after the disappearance of the nuclear membrane, are scattered evenly over the entire cell, the centromeric part of the spindle still behaves normally: the centromeres divide and the half-centromeres repel each other. In other words, the interior part of the spindle still continues to function. During this breakdown of the exterior spindle there very frequently occur all kinds of abnormal spindles, such as mono-, tri- and multipolar spindles, resulting in the origin of a varying number of nuclei in each cell. These nuclei may often be separated by cell walls.

At the same time as this inactivation of the centrosomic spindle takes place, the first signs of an abnormal behaviour also of the centromeres can be observed. Often only one or two chromosomes of one cell begin to behave abnormally. Fig. 2 a pictures such an extreme case, where just the satellited chromosome has been affected, while the rest of the chromosomes form a normal bipolar anaphase. The first visible effect of. acenaphthene on the centromeres is, in the same manner as after colchicine, a delay in the division of the centromeres. The relational spiral is completely or partly uncoiled before the division

of the centromeres (Fig. 1 *a—g*). The very typical cross-shaped c-pairs are formed in more advanced stages of the c-mitosis (Fig. 1 *h—i*). They are identical in appearance to the colchicine-induced c-pairs.

That the inactivation of the centromeres occurs gradually is de-


Fig. 1. *Allium fistulosum*, the development of the c-pairs after acenaphthene treatment, *a—g*: the uncoiling of the relational spiral, *h—i*: the cross-shaped c-pairs, *j—k*: the end stage of the c-pair formation, *l—q*: an earlier stage, where the repulsion force of the half-centromeres is still functioning. — × 3900.

monstrated, I think, by the following very typical phenomenon. Even when the division of the centromeres is delayed considerably the daughter centromeres nevertheless effect a repulsion on each other, when they eventually separate. Instead of remaining juxtaposed parallelly, as after

colchicine treatment, the chromatids now move away from each other with the centromeres in front of them (Fig. 1 *l—q*). The two-armed chromosomes are in this manner bent in the centromeric region just as at the normal anaphase (Fig. 1 *p—q*). Since, however, the exterior spindle is lacking, the half-chromosomes will move out irregularly in the plasm in all directions. Finally, it is impossible to identify which half-chromosomes originate from the same chromosome. The two half-chromosomes of the only satellited chromosome present may be recognized, however, and it may then be seen that they are very often lying in diametrically opposite corners of the cell, into which position they must have been forced by the repulsion of their centromeres.
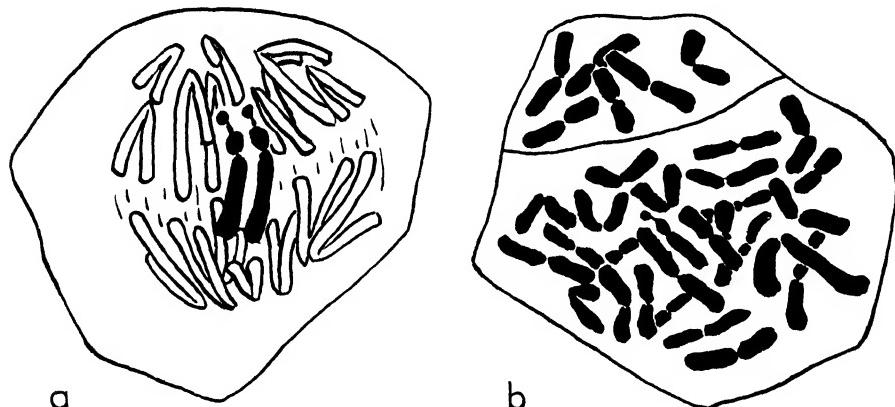
Fig. 2. *Allium fistulosum*, the break-down of the spindle after acenaphthene treatment, *a*: only one chromosome has been affected, *b*: a cell-pair originated from an abnormal anaphase. — × 2500.

As a result of the abnormalities described above a tissue will originate intermingled with cells with abnormal chromosome complements. In the most common case two cells of different size are formed at each telophase, one with more than 2x chromosomes, the other with less. It is often possible at a much later stage to recognize from the cell shape the cell-pairs that have originated in such a manner. These pairs of cells subsequently function as one cell, they enter prophase and pass mitosis together. In Fig. 2 *b* is shown such a pair of cells, which have just started a new mitosis after the preceding affected mitosis. It will be seen that the smaller cell has got 6 chromosomes at the preceding abnormal anaphase, while the larger cell has 26 chromosomes. It is evidently the intimate connection between the two cells which makes it in any way possible for a 6 chromosome cell to complete mitosis. In

pollen grains, for instance, where each cell is more definitely separated from the neighbouring cells, mitosis can never occur in a cell lacking even one single chromosome. Of course, these hypo- and hyperdiploid cells will sooner or later atrophy. And since these cells in the first week of treatment are in absolute majority, this implies a very severe strain on the vitality of the organ in question. This condition is avoided in the more instantaneous action of colchicine.

During the next period of treatment, 4—14 days, c-pairs of the same type as after colchicine treatment are found, and gradually they begin to behave like colchicine induced c-pairs, even on their separation (Fig. 1 j—k). In other words, the break-down of their centromeres is now complete. And after this the random distribution of the chromosome material of the cells ceases and pure duplications of the chromosomes are brought about. These duplications may be repeated in the same cell provided the effect of the acenaphthene is prolonged. Thus, in root tips treated for 14 days with acenaphthene there occurred numerous polyploid cells, mostly tetraploid but also octoploid and even higher polyploid cells.

The increase of the chromosome number in the cells is accompanied by an increase in the size of the cells, just as after colchicine treatment. This gives rise to the very characteristic swellings of the root tips, the so-called c-tumours. In fact, the c-tumours may be used as a rather sensitive reaction on c-mitoses. It must be remembered, however, that such tumours may be formed also by processes quite different from the c-mitoses, for instance, by treatment with growth substances (LEVAN, 1939). Due to the less complete action of acenaphthene, the c-tumours often become somewhat more extended in length than after colchicine treatment.

When the roots, after the conclusion of the treatment, are transferred into pure water, the spindle apparatus starts functioning again after a time interval of about two days, and after 3—5 days in pure water all mitoses have changed to normal. It is then seen that, in the same way as after colchicine treatment, the mitoses often have a higher chromosome number, 32 or 64, higher up in the root. In the young meristem, on the other hand, very soon normal 16 chromosome mitoses will predominate. This expresses itself macroscopically by the root tip, which grows out after the end of the treatment, rapidly narrowing down to normal thickness. But still after a considerable period in pure water the treated region may be distinguished from the younger part of the root by a more or less abrupt increase in thickness.

II. EXPERIMENTS WITH COLCHICUM.

1. *Material.* — The *Colchicum* species used, *autumnale* L., *Born-mülleri* FREYN., *byzantinum* TEN. and *speciosum* STEV., were procured from the Botanic Gardens of Lund and Copenhagen. Their normal chromosome conditions will be described elsewhere, for the present only a few data being mentioned. The somatic chromosome numbers of the
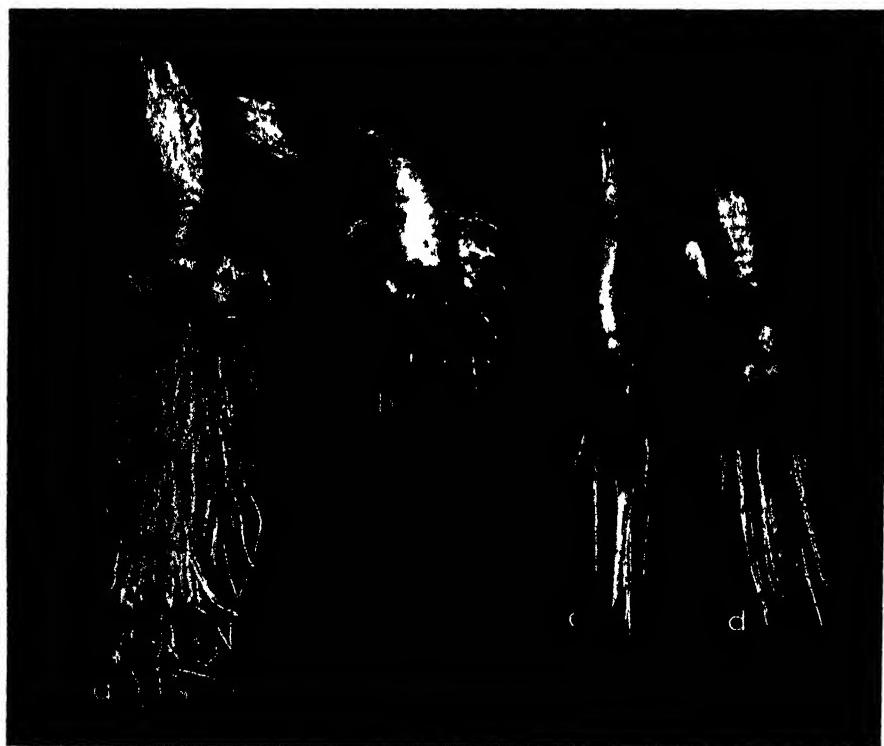

Fig. 3. *a—b: Colchicum autumnale, c—d. Colchicum Bornmülleri, b*: roots treated with acenaphthene for 20 days, *d*. roots treated with colchicine solution for 45 days *a* and *c*. controls in pure water — Photo H. OLSSON, Svalöf.

Colchicum forms studied so far are situated between 38 and 54. Within the species very considerable differences in chromosome size occur, the longest chromosomes of each idiogram being several times longer than the shortest ones. The absolute length of the chromosomes varies between 0,8 and 6 μ. Most of the chromosomes have submedially located centromeres, but some have their centromeres subterminally—terminally located. .

The treatments were made to a great extent in an unheated green-

house during October 1939, and the temperature of the greenhouse was often rather low (about $+ 2°$ C.). Root tips were fixed in NAVASHIN, which usually gave very good fixations. A great number of mitoses were present in the roots (sometimes as many as 500 mitoses in one root) and the chromosomes showed very clear centromeric constrictions.

2. *Colchicine treatments.* — Bulbs of the four *Colchicum* species used were placed in colchicine solutions of the concentrations 0,01, 0,1 and 1 %. From 6 hours to 15 days after the beginning of the treatment root tips were fixed at different intervals. And after this time some of the bulbs were left for one month in the solutions. During all this
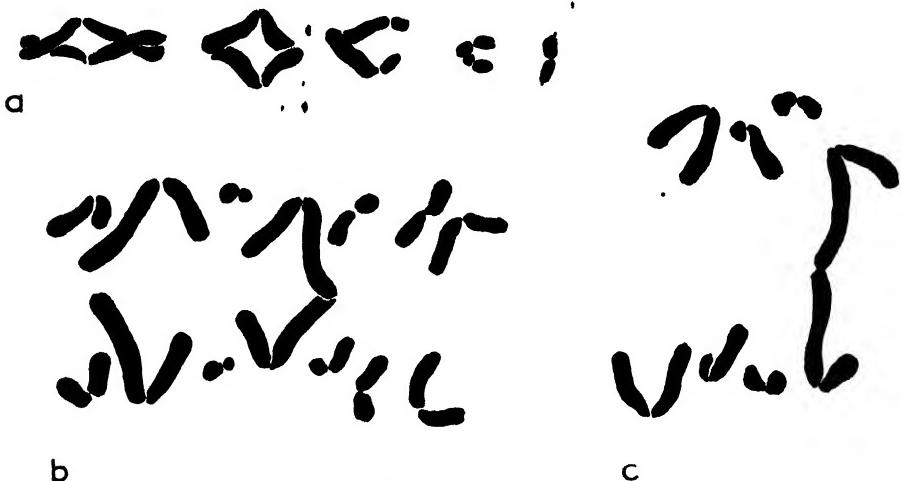
Fig. 4. *Colchicum autumnale*, anaphase chromosomes from a root tip treated for 15 days with 1 % colchicine solution. — $\times 5000$.

time no signs of c-tumours were observed. In *Allium* the formation of c-tumours is quite evident already after a treatment of one day. Fig. 3 *d* shows a *Colchicum* bulb, which was allowed to grow for $1 \frac{1}{2}$ month in a 1 % colchicine solution, while the bulb pictured in Fig. 3 *c* was left for the same time in pure water. As seen, the colchicine-treated plant has at least as long and vigorous roots as the control plant. As a matter of fact the roots continued to grow rapidly in length all through the period in colchicine.

The cytological study showed no differences whatever in the number and course of the mitoses of the treated plants, as compared with the controls. Even after 15 days in 1 % colchicine no signs of c-mitoses could be detected, and the anaphases took place absolutely normally

(Fig. 4 *a—c*). Thus it may be said that the spindle of *Colchicum* is entirely immune to colchicine.

    3. *Acenaphthene treatments.* — The 4 species employed all responded very strongly to acenaphthene. The appearance of the macroscopic c-tumour reaction is seen from Fig. 3 *b*, which pictures a bulb, the roots of which had grown for 20 days dusted with acenaphthene crystals. The difference from the control plant in Fig. 3 *a* is striking.

    The cytological study of the effect of acenaphthene on the *Colchicum* chromosomes was rendered difficult by the high number of chromosomes present and their small size. It is demonstrated beyond doubt, however, ·that acenaphthene causes real c-mitoses in *Colchicum* as well as in *Allium*.

TABLE 1. *The course of the acenaphthene effect.*

Species	Period of treatment												
	½ day		1		2		3		4		6		8	
	n[1]	c[1]	n	c	n	c	n	c	n	c	n	c	n	c
Colchicum autumnale..............		+		+	+	(+)	+		+	(+)	+			+
Colchicum Bornmülleri	+	(+)			+	(+)	+	(+)	+	(+)	+	+	(+)	+
Colchicum byzantinum			+		+		+		+		.+			
Colchicum speciosum		+	+		+				+					+

In Table 1 is given a survey of ~~the~~ acenaphthene action on the *Colchicum* mitoses. *Colchicum* seemed to be somewhat more sensitive to acenaphthene than *Allium*. In some cases a number of c-mitoses are found in the treated tissues already after 12 hours. It must be pointed out, however, that the early c-mitoses sometimes looked somewhat abnormal. Thus the c-pairs were often collected into a more or less compact ball in the centre of the cell, and not until after 2—3 days did they begin to assume a more normal appearance. In such cases the acenaphthene treatment seemed to bring about a shock effect, which required some time to fade away. In other cases even after a treatment of 8 days there were a great number of normal mitoses present. mixed with the c-mitoses.

    [1] n = normal mitoses, c = c-mitoses.

Another feature also indicating a more sudden response to ace-naphthene in *Colchicum* than in *Allium* is the very scarce occurrence of multipolar anaphases. This gave the impression that acenaphthene acts more definitively on the spindle mechanism of *Colchicum* than on that of *Allium*.

Fig. 5 shows the different stages in the development of the c-pairs of *Colchicum*. The chromosomes are scattered disorderly after the disappearance of the nuclear membrane, and the chromatid spiral begins uncoiling (Fig. 5 *a—p*). The variation in size of the *Colchicum* chro-mosomes, together with differences in the position of their centromeres, gives a very much greater diversity in appearance to the c-pairs than is the case in *Allium*. The centromeres were seldom divided during the
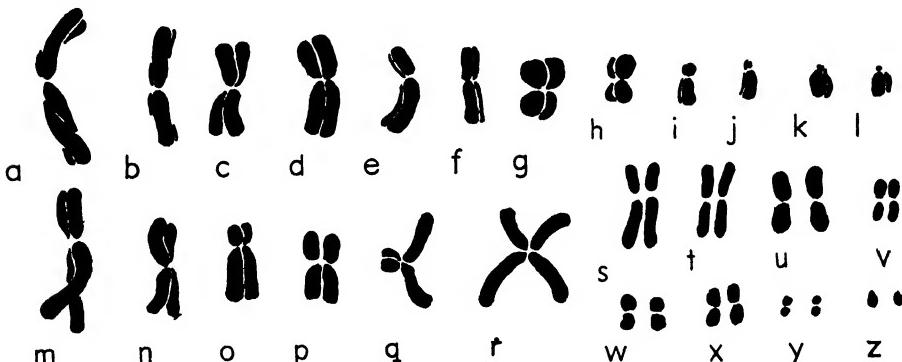
Fig. 5.   *Colchicum*, the development of the c-pairs after acenaphthene treatment. —
× 4000.

first few days of the treatments, and this accounts for the rare occur-rence of multipolar anaphases. When the centromeres on the fourth day after the beginning of the treatment started to divide the centro-meres were evidently completely inactivated, so that the half-chromo-meres did not repel each other (Fig. 5 *s—z*). On the sixth day the second and third c-mitosis in the same cell began to appear. The chromosome numbers could then, without any difficulty be estimated at about 80 and 160 respectively.

The period of spindle recovery could in some cases be followed in detail. After a treatment of four days and a period of one or two days in water single normal mitoses were found among numerous c-mitoses. After a further 5 or 10 days in pure water the same plant showed normal diploid mitoses in the meristem and solitary normal tetraploid mitoses more distant from the tip.

In another case the treatment was continued for 8 days, the recovery being thus controlled after 3, 5 and 7 days in water. In these roots a very high percentage of tetraploid cells and also some octoploid cells were met with. In the cortical cells at a rather long distance from the tip numerous abnormalities occurred even after 7 days in water. One of the most common abnormalities was the occurrence of multinucleate cells, in which the different nuclei entered mitosis simultaneously. These mitoses in the same cell took place without disturbing each other. They often resembled mitoses of the tapetum

TABLE 2. *The occurrence of polyploid mitoses in two roots of Colchicum.*

Distance from the tip	Slide 18142 (accn. 8 days + water 3 days)				Slide 18144 (accn. 8 days + water 5 days)	
	2x	4x	8x	2 or more mitoses in one cell	2x	4x
0—99 μ	93	1	—		143	7
100—199	120	7	—	—	157	23
200—299	104	2		1	72	13
300—399	95	5	—	4	43	29
400—499	66	5	1	6	—	—
500—599	20	2	1	8	—	—
Total	498	22	2	19	415	72

cells. And if two equally-sized nuclei were dividing in one cell the similarity to the second meiotic division was striking. The spindles of the different mitoses might then be orientated either side by side or behind each other in one row.

In some roots the different types of mitoses could be counted section by section. Table 2 gives two instances of such counts. As has previously been demonstrated in *Allium*, it was found also in *Colchicum* that the frequency of polyploid cells increases the greater the distance is from the tip meristem. Thus, as might be expected, the diploid cells of *Colchicum* are superior in the competition with polyploid cells within the mixoploid tissue. Nevertheless, in certain roots whole sectors were found to remain tetraploid during some time after the end of the treatment.

III. DISCUSSION.

My experiments confirm to a great extent the opinion of KOSTOFF that in its effects on mitosis acenaphthene is comparable to colchicine. The negative results with acenaphthene, which have sometimes been reported, must be due either to a special immunity of the experimental plants employed or, more likely, to an erroneous technique. The latter explanation has been suggested by KOSTOFF (1938 b). Differences in the effect of acenaphthene occur, however, in identical material as compared with the effect of colchicine. Thus, acenaphthene acts decidedly more slowly in *Allium* than colchicine. It takes perhaps 1000 times longer to obtain the maximal effect with acenaphthene than with colchicine. The reason of this may be found in the very low degree of solubility of acenaphthene in water, which necessarily retards its absorption in the cell sap. If this is the case, perhaps the external parts of the tissue should be affected first, and this could not be demonstrated in my experiments. The affected cells were found intermingled among normal cells all through the tissue.

On the other hand, the similarity of action between acenaphthene and colchicine is striking. And the final stage of the effect, the completely formed c-pairs, is identical in both cases. The acenaphthene effect, as it appears in *Allium*, may consequently be regarded as an ultra-rapid exposure of colchicine action. And this condition involves, as has been pointed out earlier, a great advantage for the detailed study of the course of the chromosome disturbances. The course of the c-mitosis becomes extremely fractionated.

Great as the advantage of this condition is theoretically, it is equally disadvantageous from a practical viewpoint, when it comes to inducing polyploidy. KOSTOFF emphasizes the lack of poisonous effect of acenaphthene as an advantage, compared with colchicine. This advantage is unfortunately diminished by the slowness in action of the acenaphthene, which must cause a predisposition to the origin of aneuploid cells. Such cells are of course under all conditions formed during the recovery stage. Colchicine, too, gives a slow and fractionated recovery process, during which all kinds of spindle disturbances are found.

It may be mentioned, however, that these conditions have been studied by me in detail only in *Allium*. It is therefore probable that other plants may exhibit another reaction, and will perhaps be to the advantage of acenaphthene in inducing polyploidy. This is of course the case under such special conditions as in *Colchicum*, which is immune

to colchicine.   According to BLAKESLEE (l. c.), the fungi are immune to
colchicine.   In other cases perhaps organisms are found to be hyper-
sensitive to colchicine, so that even small doses cause lethality.   Here,
too, the less poisonous acenaphthene will be appropriate.   It thus
follows that from a practical viewpoint it must be important that com-
parative treatments with different accessible substances are carried out
in order to make sure in each case which substance is the most suitable
one for the kind of organism under examination.

A very important branch of the intense study of the course of the
c-mitosis and the c-meiosis, which has been going on since the discovery
of the polyploidy-inducing ability of colchicine, is the analysis of differ-
ent problems connected with the mechanics of the cell divisions.   The
possibility of temporarily inactivating the spindle apparatus, affords a
valuable opportunity of an experimental approach to the forces governing
the normal course of mitosis and meiosis.   And I think that the use
of acenaphthene may be of still greater importance than colchicine in
this field.   Thus the differential reaction of the exterior and interior
spindle could be very easily studied during the acenaphthene treatment.
These two parts of the spindle, conveniently referred to as the centro-
somic and centromeric parts, are usually inactivated simultaneously by
colchicine.   It was only when concentrations just above the threshold
value were used (in *Allium* about 0,005 %) that the exterior spindle was
regularly inactivated before the interior spindle.   Acenaphthene conse-
quently acts as a very dilute colchicine solution, which is in agreement
with its very slight solubility in water.

Thus the complex nature of the spindle mechanism is more clearly
exhibited by acenaphthene than by colchicine.   Beside the difference
between the centrosomic and centromeric part of the spindle the centro-
meric function is further divided into two partial functions: one re-
productive function, which brings about the division of the centromere,
and one repelling function usually regarded as the internal spindle.   The
reproductive function is affected by acenaphthene in such a manner
that a remarkable delay in the division of the centromeres is brought
about, but this function is never arrested completely.   The repelling
function, on the other hand, is gradually eliminated during the progress
of the acenaphthene effect, so that the two daughter chromosomes of
each c-pair will eventually remain parallel beside each other.

It is impossible at present to decide if the effect of acenaphthene is
really identical with the effect of colchicine.   The experiments with
Colchicum show that the same spindle apparatus can be immune to one

of these substances and susceptible to the other. The possibility touched upon in the introduction, that colchicine and acenaphthene may act upon two different part-functions, both necessary for a normal spindle function, certainly diminishes in probability at the same time as the course of the c-mitoses caused by the two different substances are in detail similar morphologically to each other. A second alternative is that the effect of colchicine and acenaphthene are identical, the observed differences in their action being due just to differences in solubility, in reactivity etc. This second alternative necessitates the assumption of the presence of an antivenom in *Colchicum* (cf. BLAKESLEE, l. c.), which inactivates the colchicine in the cells, before the colchicine action is brought about. The genus *Colchicum* must necessarily have aquired the faculty of destroying colchicine before the faculty of producing colchicine.

SUMMARY.

The effect of acenaphthene and colchicine is studied on root mitoses of *Allium* and *Colchicum*.

In *Allium* acenaphthene is found to bring about the same deviation from normal mitosis as colchicine. The slower and less complete action of acenaphthene, as compared with colchicine, may be ascribed to its lower degree of solubility in water.

The differential reaction of one exterior and one interior part of the spindle apparatus already observed after colchicine treatment is made more evident after acenaphthene treatment.

Colchicum is shown to be entirely immune to colchicine, but highly susceptible to acenaphthene, which causes regular c-mitoses in *Colchicum*, giving rise to tetraploid and octoploid cells and sectors.

Svalöf, 14th November 1939.

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ZUR GENETIK VON PHASEOLUS VULGARIS

XVI. WEITERE BEITRÄGE ZUR VERERBUNG DER TEILFARBIGKEIT

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(With a Summary in English)

VOR einigen Jahren habe ich (LAMPRECHT, 1934) eine Arbeit über die Vererbung der Teilfarbigkeit der Testa bei *Phaseolus vulgaris* veröffentlicht, in der teils die bis dahin auf diesem Gebiet bekannten Resultate zusammenfassend besprochen, teils die Spaltungsergebnisse in F_2 von sieben Kreuzungen mitgeteilt wurden. Das bis dahin Bekannte und die Ergebnisse dieser Arbeit können etwa folgendermassen kurz zusammengefasst werden.

Für die Ausbildung von Teilfarbigkeit ist vor allem Rezessivität in den zwei Grundgenen, *T* und *E*, erforderlich. Alle *TE*-Pflanzen sind ganzfarbig. Ausserdem gibt es mehrere Teilfarbigkeitsgene, die je für sich, wie auch zusammen, die Färbung verschiedener Gebiete der Testa bedingen dürften. Zwischen diesen Genen besteht Komplexwirkung. Welches Aussehen Samen haben, die in allen Teilfarbigkeitsgenen rezessiv sind, ist bisher unbekannt. Es wäre demnach denkbar, dass die Grundgene allein schon einen gewissen Typus von Teilfarbigkeit bedingen könnten.

In der genannten Arbeit wurden 17 erblich verschiedene Typen von Teilfarbigkeit beschrieben und abgebildet. Die l. c. eingeführte Bezeichnung der Typen erfolgt mit lateinischen Namen, die die Gestalt der Zeichnung auf der Samenschale angeben. So z. B. *bipunctata* = mit zwei kleinen Flecken, *virgata* = mit Streifen, *arcus* = mit Bogen, *virgarcus* = mit Streifen und Bogen, *sellatus* = Sattel u. s. w. (vgl. die weiter unten folgenden Abbildungen). Im ganzen waren schon damals etwa 22 verschiedene Typen von Teilfarbigkeit bekannt, weshalb angenommen wurde, dass vier, wahrscheinlicher fünf, verschiedene Gene für Teilfarbigkeit bestehen.

Die Aufspaltung in den mitgeteilten Kreuzungen war, ausgenommen in einer Kreuzung, Nr. 32, unklar. Die erhaltenen Spaltungszahlen für die verschiedenen teilfarbigen Typen liessen sich nicht durch die bekannten einfacheren Schemas für 2- oder 3-Genenspaltung erklären.

Zwei Ursachen dürften hierzu wesentlich beigetragen haben. Teils waren alle Kreuzungen zwischen ganzfarbigen und teilfarbigen Linien ausgeführt, wobei sich herausstellte, dass die meisten ganzfarbigen in der genotypischen Konstitution für Teilfarbigkeit Unterschiede in mehreren Genen aufwiesen. Teils war die Klassifikation der teilfarbigen Typen in gewisser Hinsicht unsicher. Es gab allerdings kaum ganz kontinuierliche Übergänge zwischen den weniger gefärbten Typen — zwischen *unipunctata* und *virgarcus* liegend — aber bei nicht wenigen von diesen war ein gewisser Teil der Teilfarbigkeit häufig schwach ausgebildet. Dies wurde damals als durch modifikative Einflüsse verursacht aufgefasst. Wie unten gezeigt wird, beruhen diese Erscheinungen in der Hauptsache auf Heterozygotie in den Genen für Teilfarbigkeit.

Genpaare für Teilfarbigkeit sind bisher erst zwei in ihrem Effekt näher charakterisiert. Das eine Genpaar ist $Z-z$ und bezieht sich auf die drei Typen sellatus, ZZ, Piebald, Zz, und virgarcus, zz. Belege für diese Spaltung stammen von E. v. TSCHERMAK (1912), SURFACE (1916) sowie SAX und MCPHEE (1923). Das zweite Genpaar wurde vom Verfasser (LAMPRECHT, 1934) aufgestellt und abgeleitet von *bipunctata* mit $Bip-bip$ bezeichnet. Es ist eines der Gene, die für die Spaltung *virgarcus* (dominant) : *bipunctata* (rezessiv) (mit Zwischentypen) verantwortlich sind (vgl. unten Fig. 1 und 2).

Seither hat F. SCHREIBER (1934) eine Arbeit veröffentlicht, laut der es einen dominanten »Löschfaktor» für Teilfarbigkeit gibt. Dieser sollte also die Ausbildung von Teilfarbigkeit ganz verhindern, sodass anstatt teilfarbigen, reinweisse Samen resultieren. Selbst habe ich auch in mehreren Kreuzungen (siehe l. c.) die Ausspaltung von weisssamigen aus teilfarbigen Pflanzen beobachtet, ohne jedoch hierbei ein über Teilfarbigkeit allgemein dominierendes Gen gefunden zu haben.

KREUZUNG BIPUNCTATA- × VIRGATA-TYPUS.

Diese Kreuzung, Nr. 253, wurde ausgeführt zwischen Linie 5 vom *bipunctata*-Typus, herstammend aus der französischen Brechbohnensorte *Incomparable*, und Linie 88 vom *virgata*-Typus, ausgelesen aus der englischen Brechbohnensorte *Early Giant*. Der *bipunctata*-Typus ist in Fig. 1 links, der *virgata*-Typus in Fig. 1 rechts abgebildet. Beide Typen zeigen eine gewisse, aber doch nur relativ geringe Variation (vgl. Fig. 9 in LAMPRECHT, 1934, wo 3 Samen abgebildet sind, die etwa die Variationsbreite von L. 5 angeben, sowie Fig. 19 l. c., in der der mittlere und rechte Samen dem *virgata*-Typus von L. 88 entsprechen).

Die auf den F_1-Pflanzen erhaltenen Samen zeigten die Zeichnung des mittleren Samens in Fig. 1. Wie ersichtlich hat dieser die beiden für den *bipunctata*-Typus charakteristischen zwei Flecken gleich entwickelt wie bei der Elternlinie, d. h. sie zeigten die gleiche Variationsbreite wie bei dieser. Anders verhält es sich mit dem auf der Mikropylenseite befindlichen Streifen des *virgata*-Samens. Dieser ist hier nicht voll ausgebildet sondern nur durch Punkte angegeben, die allerdings mitunter mehr oder weniger miteinander verfliessen können. Man konnte schon unter den auf F_1 erhaltenen Samen solche antreffen, in denen es schwer erschien sie sicher von schwach ausgebildeten *virgata* der Elternlinie zu unterscheiden.
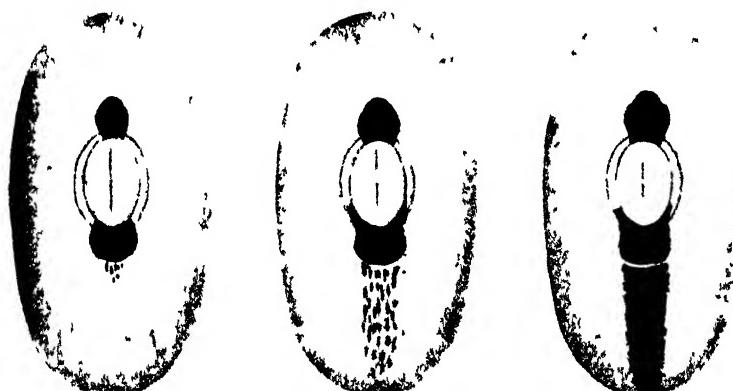
Fig 1   Links ein Same vom *bipunctata*-Typus, *bip arc*, rechts ein solcher vom *virgata*-Typus, *bip Arc*   Der mittlere Same entspricht der heterozygoten Konstitution *bipbip Arcarc* (F_1 von Kreuzung Nr. 253).

In der zweiten Generation wurden 600 Samen gesät, die im ganzen 537 voll reifende Pflanzen entwickelten. Die Klassifikation der *bipunctata*-Samen verursachte keinerlei Schwierigkeit, sie waren leicht von den übrigen zu scheiden. Bei den *virgata*-Samen dagegen erschien es schwierig eine bestimmte Grenze zu ziehen zwischen voll ausgebildeten *virgata*-Typen (= Elternlinie 88) und solchen mit nur durch Punkten angedeutetem Streifen (vgl. LAMPRECHT, 1934, Fig. 19, linker und mittlerer Samen). Die zwischen den letztgenannten beiden Typen gezogene Grenze ist also nicht scharf. Für die drei Typen in F_2 ergaben sich folgende Zahlen:

Gefunden: 132   *bipunctata* : 294   *schwach virgata* : 111   *typisch virgata*
Erwartet: 134,25   »   : 268,50   »   » : 134,25   »   »
D/m für
1 : 2 : 1 =   0,22   2,20   2,31

Es besteht kein Zweifel, dass es sich hier um eine monohybride Spaltung nach dem *Zea*-Typus handelt, bei der die Abgrenzung der heterozygot von den homozygot *virgata* nicht ganz sicher erscheint. In diesem Gebiet gibt es Zwischentypen, deren Zugehörigkeit erst durch Untersuchung in einer weiteren Generation sichergestellt werden kann. Die F_3-Generation hat dies auch bestätigt. Es wurden insgesamt 22 Familien untersucht. Sämtliche *bipunctata*-Typen, 6 Familien mit zusammen 109 Individuen, haben nur wiederum solche Nachkommen gegeben. Und das Gleiche war mit den Nachkommen nach typischen *virgata* der Fall, 5 Familien mit 92 Pflanzen verblieben konstant *virgata*. Von den 11 Familien, die nach *schwach virgata* gebaut wurden, waren dagegen 2 konstant *virgata*, jetzt mit deutlich ausgebildetem *virgata*-Streifen,
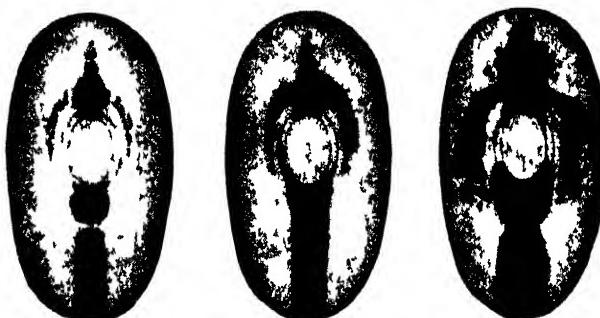
Fig 2   Drei Samen vom *virgarcus*-Typus der Linie 57 aus der Sorte Goldregen Formel *Bip Arc*   Der linke Same hat ungewöhnlich schwach ausgebildeten *arcus*

während die übrigen 9 Familien im Verhältnis 51 *bipunctata* : 92 *schwach virgata* : 34 *typisch virgata* spalteten. Hier liegt also offenbar wiederum dasselbe 1 : 2 : 1-Verhältnis wie in F_2 vor.

Für den Unterschied zwischen *virgata* und *bipunctata* ist demnach ein Gen verantwortlich zu machen, das in seiner dominanten Form die *virgata*-, in seiner rezessiven die *bipunctata*-Zeichnung bedingt. Ein Symbol für dieses Gen wird bei Besprechung der nächsten Kreuzung eingeführt werden.

KREUZUNG BIPUNCTATA- × VIRGARCUS-TYPUS.

Diese Kreuzung, Nr. 251, wurde ausgeführt zwischen Linie 5 (wie in voriger Kreuzung) und Linie 57, aus der deutschen Wachsbohnensorte Goldregen, die den *virgarcus*-Typus repräsentiert. Die Samen des letzteren zeigen die in Fig. 2 wiedergegebene Zeichnung und Variation.

Die auf F_1 erhaltenen Samen zeigten die Zeichnung des rechten

Samens in Fig. 4. Die Ausbildung des *virgata*-Streifens sowie des vom oberen Fleck nach beiden Seiten des Hilums ausgehenden Bogens variierte in der Stärke, erreichte aber niemals diejenige der Elternlinie
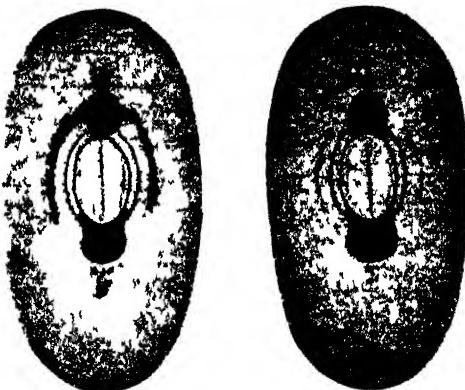

Fig 3   Links *arcus*-Typus homozygot und gut ausgebildet   *BipBip arcarc*, rechts *arcus* Typus heterozygot   *Bipbip arcarc*

57 (Fig. 2). Mitunter waren diese beiden Abzeichen schwach ausgebildet, aber doch stets noch erkennbar.

Die zweite Generation bestand aus 552 Individuen (gesät 600
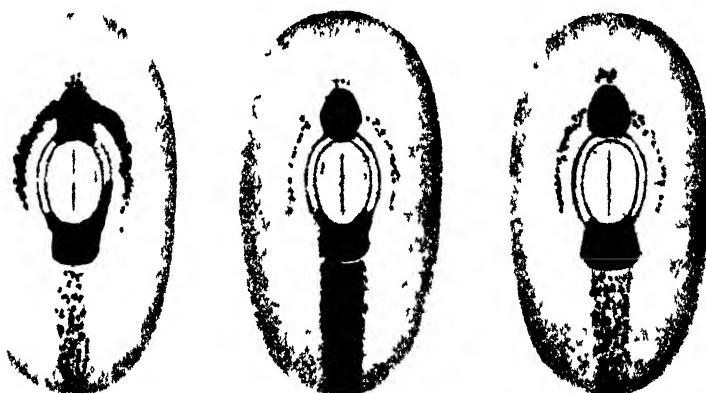
Fig 4   Drei verschieden heterozygote *virgarcus*-Typen; links *virgata*-heterozygot· *BipBip Arcarc*, Mitte *arcus*-heterozygot· *Bipbip ArcArc*; rechts *virgata*- und *arcus*-heterozygot *Bipbip Arcarc*.

Samen). Die auf dieser erhaltenen Samen wurden in neun verschiedene Gruppen eingeteilt, entsprechend den neun verschiedenen, möglichen Genkombinationen einer bifaktoriellen Spaltung. Die Farbenverteilungen auf der Testa und die diesen entsprechenden Genkonstitutionen ergeben sich aus den Fig. 1—4.

Das eine der beiden hier wirksamen Gene ist das bereits früher bekannte Gen *Bip* (LAMPRECHT, 1934). Das zweite bezeichne ich mit *Arc*, abgeleitet von *arcus* = Bogen, Fig. 3 links. Diese, der sog. *arcus*-Typus, der in *arc* doppeltrezessiv ist, wird also durch einen farbigen Bogen zu beiden Seiten des Hilums charakterisiert. Zwischen Teilfarbigkeitstypen und diesen beiden Genen ergeben sich dann folgende Beziehungen.

BipBip ArcArc = *virgarcus*-Typus homozygot, sowohl Bogen wie Streifen stark ausgebildet; Fig. 2.

BipBip Arcarc = *virgarcus*-Typus *Arc* heterozygot, Bogen gut ausgebildet, Streifen nur in mehr oder weniger deutlichen Punkten angedeutet; Fig. 4 links.

BipBip arcarc = *arcus*-Typus homozygot, Bogen schön ausgebildet, Streifen fehlt; Fig. 3 links.

Bipbip ArcArc = *virgarcus*-Typus *Bip*-heterozygot, Streifen stark ausgebildet, Bogen nur in Punkten angedeutet; Fig. 4 Mitte.

Bipbip Arcarc = *virgarcus*-Typus *Arc*- und *Bip*-heterozygot, sowohl Bogen wie Streifen nur in mehr oder weniger deutlichen Punkten angedeutet; Fig. 4 rechts.

Bipbip arcarc = *arcus*-Typus *Bip*-heterozygot, Bogen nur in mehr oder weniger deutlichen Punkten angedeutet (mitunter ganz fehlend), Streifen fehlt stets; Fig. 3 rechts.

bipbip ArcArc = *virgata*-Typus homozygot, Bogen fehlt, Streifen stark ausgebildet; Fig. 1 rechts.

bipbip Arcarc = *virgata*-Typus *Arc*-heterozygot, Bogen fehlt, Streifen nur in mehr oder weniger deutlichen Punkten ausgebildet; Fig. 1 Mitte.

bipbip arcarc = *bipunctata*-Typus homozygot, Bogen und Streifen fehlt, nur die beiden Flecken an Caruncula und Mikropyle sind vorhanden; Fig. 1 links.

Aus den Figuren ist ferner ersichtlich, dass auch beim *bipunctata*- und beim *arcus*-Typus einige Farbpunkte des Streifens vorkommen können, aber bei diesen Typen befinden sich diese dann nur in unmittelbarer Nähe der beiden Flecken an Caruncula und Mikropyle (vgl. Fig. 1 links und 3). Eine Verwechslung mit dem *virgata*-Typus erscheint hierdurch kaum möglich (vgl. Fig. 1 Mitte und 4 links und rechts).

Die Verteilung der 552 Pflanzen der F_2-Generation auf die oben angeführten neun Gruppen war die folgende:

		Gefunden:	Theoretisch erwartet:
virgarcus-	*BipBip ArcArc* konstant *virgarcus*	35	34,5
Gruppe	*BipBip Arcarc virgata*-spaltend	16	69,0
	Bipbip ArcArc arcus-spaltend	78	69,0
	Bipbip Arcarc virgata-u. *arcus*-spaltend	205	138,0
	S:a	334	310,5
arcus-	*BipBip arcarc* konstant *arcus*	2	34,5
Gruppe	*Bipbip arcarc arcus*-spaltend	33	69,0
	S:a	35	103,5
virgata-	*bipbip ArcArc* konstant *virgata*	14	34,5
Gruppe	*bipbip Arcarc virgata*-spaltend	93	69,0
	S:a	107	103,5
bipunctata-			
Gruppe	*bipbip arcarc* konstant *bipunctata* S:a	76	34,5

Bei Betrachtung dieser Zahlen kann folgendes festgestellt werden. Die gefundenen Individuensummen für die beiden Gruppen *virgarcus* und *virgata* zeigen mit den bei Zweigenenspaltung erwarteten gute Übereinstimmung. In der *arcus*-Gruppe besteht dagegen ein starkes Defizit und in der *bipunctata*-Gruppe ein etwa dementsprechender Überschuss. Vereinigt man diese beiden Gruppen, so resultieren folgende Zahlen.

Gefunden: 334 *virgarcus* : 107 *virgata* : 111 *arcus-bipunctata*
Erwartet: 310,5   »   : 103,5   »   : 138,0   »   »
D/m für
9 : 3 : 4 =   2,02   0,19   2,66

Es handelt sich also zweifellos um eine bifaktorielle Spaltung in den beiden Genpaaren *Bip—bip* und *Arc—arc*, nur lässt die Abgrenzung der verschiedenen heterozygoten Gruppen zu wünschen übrig. Innerhalb jeder der beiden Gruppen *virgarcus* und *virgata* finden wir wiederum dieselbe Unsicherheit bei der Abgrenzung des homozygoten gegen den heterozygoten Streifen wie in der vorigen Kreuzung, nur hier in noch stärkerem Masse. Ferner ist die Ausbildung des *arcus* meistens schlecht.

Nur 2 Individuen wurden als homozygot *arcus* und 33 als heterozygot klassifiziert. Der Rest, etwa 60 Individuen, kam in die *bipunctata*-Gruppe. Dies bewies die dritte Generation, in der aus *bipunctata* sowohl hetero- wie homozygote *arcus*-Typen ausspalteten. Da die Ausbildung des *arcus* sowohl in Linien wie in anderen Kreuzungen jedoch eine gute sein konnte, erscheint es möglich, dass in Kr. 251 noch ein Gen wirkt, das die schwache und unsichere Ausbildung dieses Typus bedingt. Wahrscheinlich handelt es sich hierbei um ein Teilfarbigkeitsgen, das beide Elternlinien in gleicher Allelenform enthalten.

KREUZUNG BIPUNCTATA- × MAJOR-TYPUS.

Diese Kreuzung, Nr. 174, wurde ausgeführt zwischen L. 5 (wie in voriger Kreuzung) und L. 6 aus der französischen Brechbohnensorte

TABELLE 1. *Die Aufspaltung des Bastards Arcarc Bipbip Diffdiff in*
F₂ von Kreuzung Nr. 174.

Genenspaltung			Teilfarbig-keitstypus	Individuenanzahl		D/m
				Gefunden	Erwartet	
F_1:	48 *Arc* {36 *Bip*	{ 27 *Diff*	*virgarcus*	238	231,19	0,59
		9 *diff*	*maximus*	66	77,06	1,36
	12 *bip*	{ 9 *Diff*	*virgata*	60	77,06	2,10
		3 *diff*	*major*	29	25,69	0,67
	16 *arc* {12 *Bip* $\left(\frac{Diff}{diff}\right)$		*arcus*	155	137,00	1,77
	4 *bip* $\left(\frac{Diff}{diff}\right)$		*bipunctata*			

Très nain précoce. Letztere repräsentiert den *major*-Typus; siehe Fig. 5. Die auf der ersten Generation erhaltenen Samen zeigten die in Fig. 6 wiedergegebene Zeichnung. Wie ersichtlich stehen diese Samen dem *virgarcus*-Typus nahe, doch mit etwas stärkerer Ausbildung des *arcus* und zweier Lappen an den Seiten der Mikropyle. In den beiden vorigen Kreuzungen zeigte sich, dass Heterozygotie in den beiden Genen für Teilfarbigkeit *Arc* und *Bip* intermediäre Typen ergab. Dasselbe scheint auch hier der Fall zu sein, jedoch mit einer starken Annäherung an den Typus init geringerer Ausbreitung von Farbe auf der Testa. Die grössere Ausbreitung der Farbe, wie sie der eine Elter (Linie 6) zeigt, scheint

daher durch Rezessivität in einem weiteren Teilfarbigkeitsgen bedingt zu
werden.

In der zweiten Generation wurden 600 Samen gesät, die 548 samen-
tragende Pflanzen entwickelten. Diese konnten hinsichtlich Teilfarbig-

Fig 5   Drei Samen vom *major*-Typus aus Linie 6, Très nain précoce   Der eine Elter
von Kreuzung Nr 174

keit hauptsächlich in folgende 6 verschiedene Typen eingeteilt werden:
bipunctata, arcus, virgata, virgarcus, maximus und *major* (vgl. Fig. 1—5
und 7). Die fur diese sechs Typen gefundenen und theoretisch erwar-
teten Individuenanzahlen sind in Tabelle 1 zusammengestellt. Aus dieser

Fig 6   Die Zeichnung der auf *F₁* von Kreuzung Nr 174 erhaltenen Samen, Formel
Bipbip Arcarc Diffdiff

ist vor allem ersichtlich, dass teils eine Spaltung in den beiden in voriger
Kreuzung behandelten Genen *Arc* und *Bip*, teils eine Spaltung in einem
weiteren Gen stattgefunden hat, das als *Diff* bezeichnet wurde. Da nun
die eine Elternlinie, Nr 5, vom *bipunctata*-Typus in *Arc* und *Bip* re-
zessiv ist (vgl. die vorige Kreuzung), kann auf Grund der gefundenen

Spaltung geschlossen werden, dass der durch die andere Elternlinie, Nr. 6, repräsentierte *major*-Typus in diesen beiden Genen dominant sein muss. Ferner muss sich der *major*-Typus in wenigstens noch einem weiteren Gen vom *bipunctata*-Typus unterscheiden. Dieses neue Gen bedingt laut den Spaltungsresultaten in rezessiver Form eine Ausbreitung der Farbe auf der Testa. Mit Hinblick hierauf wurde es mit dem Symbol *diff*, abgeleitet von *diffundere* = ausbreiten, belegt.

In den beiden Genpaaren *Arc—arc* und *Bip—bip* erhalten wir die gleiche Spaltung wie in der vorigen Kreuzung, nämlich dem Verhältnis 9 : 3 : 4 entsprechend, da auch hier wiederum die Grenze zwischen den beiden Typen *arcus* und *bipunctata* nicht scharf ist. Der *arcus* ist oft sehr schwach ausgebildet oder nicht mehr erkennbar, sodass ein Teil
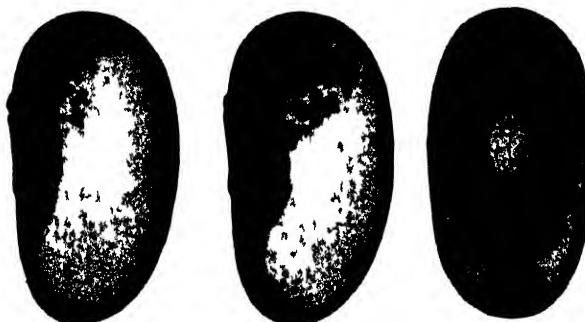
Fig 7   Drei Samen vom *maximus*-Typus

der *arcus*-Typen als *bipunctata* klassifiziert werden. Fur die Spaltung in diesen beiden Genen ergaben sich folgende Zahlen:

Gefunden: 304 *Arc Bip* : 89 *Arc bip* : 155 *arc* $\left(\begin{matrix} Bip \\ bip \end{matrix}\right)$

Erwartet: 308,25 »   » : 102,75 »   » : 137,00 »   »

D/m fur

9 : 3 : 4 =   0,37   1,51   1,77

Wie ersichtlich ist die Übereinstimmung zwischen gefundenen und theoretisch erwarteten Zahlen befriedigend. Von den *arc*-Individuen wurden 72 als *arcus* und 83 als *bipunctata* klassifiziert, was auf die vorhin erwähnte schlechte Ausbildung des *arcus* zurückzuführen ist.

Das neue Gen *diff* scheint bei Rezessivität in *arc*, wenigstens bei den hier in Frage kommenden genotypischen Konstitution, weder in dominanter noch in rezessiver Form einen Einfluss zu besitzen. In der Gruppe der *Arc*-Individuen bedingt Rezessivität in *diff* eine starke Ausbreitung

der Teilfarbigkeit. Der *virgarcus*-Typus wird durch *diff* in den *maximus*-, der *virgata*-Typus in den *major*-Typus umgewandelt. Es. kann hier die nicht häufige Erscheinung festgestellt werden, dass gewisse Gene für eine Eigenschaft (Teilfarbigkeit) diese in dominanter Form verstärken, während ein anderes Gen (*diff*) dies in rezessiver Form bedingt. D. h. *Arc* und *Bip* verursachen in dominanter Form grössere Ausbreitung der Farbe auf der Testa, *Diff* bedingt grössere Ausbreitung dagegen in rezessiver Form.

Wie schon für F_1 erwähnt worden ist, zeigen *virgarcus*-Samen bei Heterozygotie in *Diff* ein wenig grössere Verbreitung der Farbe (s. Fig. 6). Dasselbe scheint auch für den *virgata*-Typus zu gelten. Auch
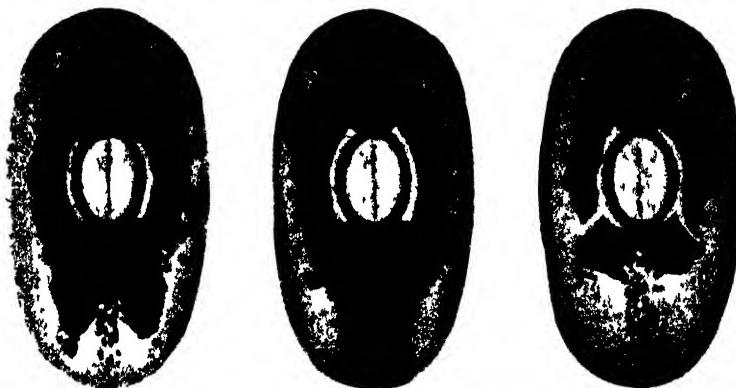
Fig 8   In Kreuzung 174 ausgespaltene Samen vom. *major*- und *maximus*-Typus, die in einem oder mehreren Genen heterozygot sind   Links *major*-Typus, der in *virgata* (*Bipbip*) und vielleicht auch in *arcus* heterozygot ist, in der Mitte ein Same mit homozygot *virgata*, aber wahrscheinlich heterozygot *arcus*   Rechts *maximus*-Same mit heterozygot *virgata* und wahrscheinlich auch *arcus*

bei den Samen vom *maximus*- und *major*-Typus, die also beide in *diff* homozygot rezessiv sind, kann Heterozygotie in *Arc* und *Bip* an der Zeichnung festgestellt werden. Als Beispiel hierfür soll Fig. 8 dienen. Wie aus dieser ersichtlich, wird die Ausbreitung der Farbe bei Heterozygotie in *Bip* offenbar vermindert. Heterozygotie in *Arc*, die den Streifen des *virgata*-Typus in Punkte auflöst, kann, wie der linke und rechte Samen in der Fig. zeigen, leicht festgestellt werden. Beim mittleren Samen geht der Streifen breit und stark um das Ende des Samens herum; er ist hier deutlich homozygot, *ArcArc*. Die durch das Gen *diff* zu beiden Seiten der Mikropyle bedingten Lappen scheinen stets deutlich ausgebildet zu werden (vgl. z. B. Fig. 8 rechter Samen).

Eine scharfe Abgrenzung aller dieser verschiedenen heterozygoten Typen gegeneinander ist ziemlich schwierig und wahrscheinlich nur

dann sicher durchzuführen, wenn Spaltung nur in einem der betreffenden Gene stattfindet.   Hierzu sind also weitere Analysen in F_3 und F_4
usw, bzw. in speziell hierauf abzielenden Kreuzungen erforderlich.

KREUZUNG VIRGARCUS- × MINOR-TYPUS.

Als *virgarcus*-Typus wurde zu dieser Kreuzung Linie 57, aus der
deutschen Wachsbohnensorte Goldregen stammend, benutzt, die auch
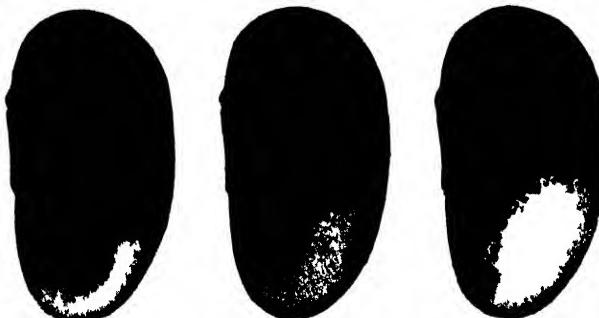
Fig 9. Drei Samen vom *minor*-Typus, Linie 106, der eine Elter von Kreuzung Nr 282

zu Kr. 251 als Elter verwendet worden ist. Der zweite Elter, Linie 106,
stammt wahrscheinlich von einer spontanen Kreuzung und wurde als

Fig. 10.   Drei Samen vom *minimus*-Typus, Linie 63

Einmischung in einer Samenprobe angetroffen. Bei Vermehrung seiner
Nachkommen zeigte sich, dass diese hinsichtlich Teilfarbigkeit konstant
waren. Die Testafarbe ist Kastanienbraun, Formel: *P C J G b V r t*. Der
minor-Typus (vgl. Fig. 9) nimmt eine intermediäre Stellung zwischen
dem *major*- und dem *minimus*-Typus ein (vgl. Fig. 10). Ohne sich von
der erblich bedingten Konstanz des *minor*-Typus überzeugt zu haben,
wäre· man am ,ehesten geneigt, denselben als eine Modifikation des
major-Typus anzusprechen, bei dem die Farbe eine etwas grössere Ver-

breitung erhalten hat.   Er steht nämlich dem *major*-Typus in der Aus-
breitung der Farbe etwas näher als dem *minimus*-Typus.

Die auf F_1 dieser Kreuzung erhaltenen Samen zeigten die in Fig. 11
wiedergegebene Zeichnung.   In der Ausbreitung der Farbe entsprachen
sie also am ehesten dem *major*-Typus, unterscheiden sich aber von
diesem bestimmt durch die zu beiden
Seiten des Streifens (*virgata*) gegen
die Mikropyle gerichteten hellen Ein-
schnitte.   Beim *major*-Typus fehlen
überdies auch die hier oberhalb
der Lappen vorhandenen hellen Ein-
schnitte.   Da der Streifen (*virgata*) voll
und stark ausgebildet ist, sollen die
Samen *ArcArc* sein.   Es wäre demnach
zu erwarten, dass der eine Elter,

Fig. 11. Auf F_1 von Kreuzung Nr.
282 erhaltener Same. Formel: *Bipbip
ArcArc Diffdiff Exexp.*

L. 106, der *minor*-Typus *ArcArc* in seiner genotypischen Konstitution
hat. F_2 hat dies auch bestätigt.

Die zweite Generation spaltete ausser in den Typen *virgata* und
virgarcus in einer Serie von Typen, die hinsichtlich Ausbreitung der
Farbe alle Übergänge vom *maximus*- bis zum *minimus*-Typus aufweist.
Eine Abgrenzung der vier Typen *maximus, major, minor* und *minimus*
sowie ihrer Heterozygoten, entsprechend ihrer genotypischen Konstitu-
tion ist daher nicht möglich gewesen.   Die folgende Übersicht über die
erhaltenen Typen ist also nur als Orientierung zu betrachten und können
die gefundenen Zahlen nicht zu einer zahlenmässigen Bearbeitung der
Spaltungsverhältnisse verwendet werden.   Es wurden gefunden:

Teilfarbigkeits-Typus	Anzahl Individuen
virgata	28
virgarcus, heterozygot in *arcus*	96
virgarcus	38
maximus, mit geringerer Farbenausbreitung	79
maximus	45
major, mit geringerer Farbenausbreitung	66
major	52
minor, mit geringerer Farbenausbreitung	41
minor	47
minimus, mit geringerer Farbenausbreitung	56
minimus	26

Summe: 574

Wenn also die vorstehend angeführten Zahlen auch zu einer exakten Analyse der stattgefundenen Spaltung ganz ungeeignet sind, so lassen sie doch in bezug auf die beteiligten Gene einen recht sicheren Schlusssatz zu. In der vorigen Kreuzung, Nr. 174, wurde ein Gen *diff* festgestellt, das in rezessiver Form den *virgata*- in den *major*-Typus und den *virgarcus*- in den *maximus*-Typus umwandelte. Dieselben Typen werden auch in vorliegender Kreuzung gefunden. Es liegt demnach Spaltung in den beiden Genen *Bip* und *Diff* vor. Es wurden aber überdies noch zwei weitere Typen von Teilfarbigkeit, nämlich *minor* und *minimus,* beobachtet. Die erbliche Konstanz dieser Typen ist schon früher festgestellt worden (LAMPRECHT, 1934); d. h. also, dass sie in homozygoter Form auftreten. In vorliegender Kreuzung muss also noch ein Gen spalten, das für die Ausbildung dieser beiden Typen verantwortlich ist und das offenbar durch den einen teilfarbigen Elter, den *minor*-Typus, eingeführt worden ist. Da die beiden teilfarbigen Typen mit grösster Ausbreitung von Farbe auf der Testa, der *minor*- und der *minimus*-Typus, in geringerer Anzahl auftreten als der *maximus*- und der *major*-Typus, sowie da der *minimus*-Typus, der die grösste Ausbreitung der Farbe aufweist, überdies am seltensten ist, ist die Wirkung des hier in Frage stehenden Gens offenbar eine dem Gen *diff* entsprechende, d. h. es bedingt gleichwie dieses in rezessiver Form grössere Ausbreitung der Farbe. Das hierfür verantwortliche Gen sei, abgeleitet von *expandere* = ausbreiten, mit dem Symbol *Exp* belegt.

Eine Abgrenzung der verschiedenen Heterozygoten gegenüber den Homozygoten in der Typenserie *maximus—major—minor—minimus* wird nur in Kreuzungen mit Spaltung in je einem einzigen Gen für Teilfarbigkeit, und auch dann vielleicht nur durch quantitative Methoden, möglich sein.

SUMMARY.

1. To increase our knowledge of the inheritance of partly colouring of the seed coat of *Phaseolus vulgaris* four crosses were studied.

2. The two types of partly colouring *bipunctata* and *virgata* (Fig. 1) differ in the gene *Arc*. The genotypical constitution of the *bipunctata*-type is *bipbip arcarc,* that of the *virgata*-type is *bipbip ArcArc.* Heterozygosity in *Arc* causes the formation of an intermediate type (Fig. 1).

3. Through yet another gene, *Bip*, in co-operation with *Arc*, the following types of partly colouring arise: *arcus, BipBip arcarc* (Fig. 3), *virgarcus, BipBip ArcArc* (Fig. 2). Heterozygosity in the gene *Bip* also causes intermediate types (Fig. 4).

4. While the two genes *Arc* and *Bip* in their *dominant* state cause the extension of colour on partly coloured types, the two new genes *Diff* and *Exp* do so in their *recessive* state. These two genes *Diff* and *Exp* are responsible for the formation of the types: *maximus, major, minor* and *minimus* (Figs. 5, 7, 9, 10). Heterozygosity in these genes causes also intermediate types (Figs. 6, 8, 11). In such cases the result will be a series of gradual transitions, as e. g. in cross No. 282 from *maximus* to *minimus*.

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ZUR GENETIK VON PHASEOLUS VULGARIS XVII—XVIII

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(With a Summary in English)

XVII. ZWEI NEUE GENE FÜR ABZEICHEN AUF DER TESTA, Punc UND Mip, SOWIE ÜBER DIE WIRKUNG VON V UND Inh.

ALS Abzeichen auf der Testa von *Phaseolus vulgaris* werden vom Hilumrand oder dessen unmittelbarer Nähe ausgehende, lokalisierte Zeichnungen aufgefasst. Von solchen sind bisher vier auf ihre Vererbung hin untersucht worden. Es sind dies Carunculastrich, Mikropylenstreifen, Corona und Margo.

Der *Carunculastrich* bildet eine Fortsetzung der Spitze der Caruncula nach aussen. Er stellt einen farbigen Strich in dunklerer Farbe als die Testa dar und ist an seinem Ende gewöhnlich deutlich gabelförmig gespalten (vgl. Fig. 1). Dieses Abzeichen wird durch das dominante Gen *Ca* bedingt (LAMPRECHT, 1932 b). Die Farbe des Carunculastriches liegt in den Pallisadenzellen.

Der *Mikropylenstreifen* bildet einen etwa 0,5—0,7 mm breiten, gewöhnlich graulichen Streifen, der, wie der Namen andeutet, auf der Mikropylenseite gleich ausserhalb des Hilumrandes seinen Ausgang nimmt und bis zum oder über das schmale Ende des Samens fortsetzt (vgl. Fig. 1). Er stellt eine rezessive Eigenschaft dar und wird durch das Gen *mi* bedingt. Er scheint nur bei Anwesenheit von *J* realisiert werden zu können (LAMPRECHT, 1932 b).

Die *Corona* bildet einen farbigen, den Hilumrand aussen umfassenden Ring, der an der Stelle der Caruncula, soweit bisher beobachtet, stets unterbrochen ist. Die Corona kann vom Hilumrand zuweilen durch einen feinen helleren Rand getrennt sein oder sie schliesst — häufiger — direkt an den Hilumrand an (vgl. Fig. 2). Die Farbe der Corona ist von der genotypischen Konstitution für die Testafarbe abhängig. Gewisse Testafarben scheinen also nur mit bestimmter Coronafarbe vorkommen zu können. Die Ausbildung der Corona wird durch das rezessive Gen *cor* bedingt. *Cor cor*-Samen haben schwach angedeutete Corona (LAMPRECHT, 1934).

Die *Margo* schliesslich bildet einen breiten, farbigen Rand, eine sog. Bräme aussen rund um Corona bzw. Hilumrand. Die Abgrenzung der Margo gegen die Farbe der übrigen Testa ist, zum Unterschied gegen die vorher genannten drei Abzeichen, nicht scharf sondern diffus (vgl.
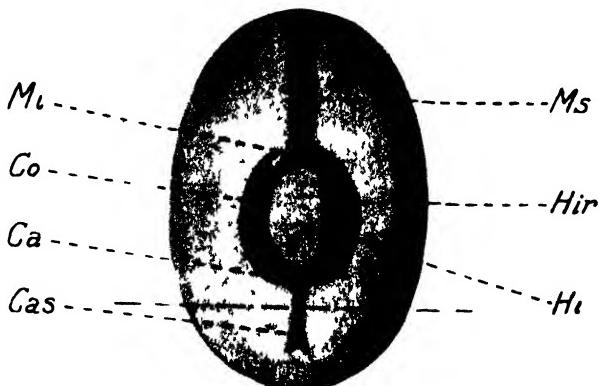

Fig 1   Abbildung einer Bohne, verschiedene Elemente bzw. Zeichnungen der Testa veranschaulichend   Hi = Hilum, Hir = Hilumrand, Ca = Caruncula, Cas = Carunculastrich, Mi — Mikropyle, Ms = Mikropylenstreifen, Co = Corona.

Fig. 3). Die Ausbildung der Margo wird durch das rezessive Gen *mar* verursacht. Die Margo kann in ganz verschiedenen Farben auftreten. Eine Reihe von solchen Fällen liegen nunmehr eingehend genetisch analysiert vor (LAMPRECHT, 1933 und 1939). Es zeigte sich, dass die
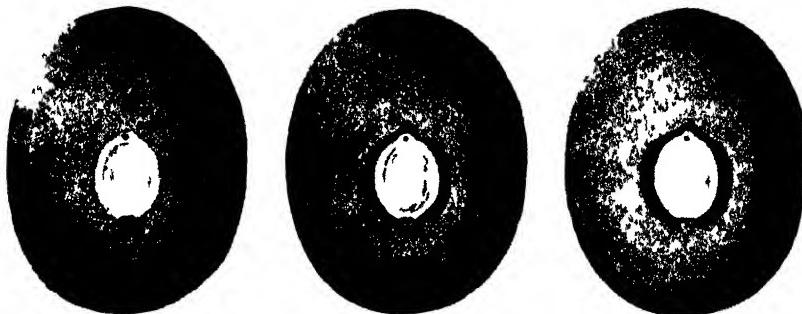
Fig 2   Drei Samen der Testafarbe Geschwefeltes Weiss   Linker Same ohne Corona, *CorCor*, rechter Same mit starker Corona, *corcor*, mittlerer Same mit schwach angedeutete Corona, *Corcor*

Margo nur bei Rezessivität im Gen *J* ausgebildet werden kann. Die Margo erscheint nämlich stets in der Testafarbe, die durch Umwandlung von *j* in *J* erhalten wird. Speckweisse Samen, *P G*, bekommen also durch *mar* eine Margo in der *P G J* entsprechenden Farbe, d. i. Maisgelb; und Gleiches gilt in allen bisher studierten Fällen. Das Gen

mar bedingt also nichts anderes, als lokale Umwandlung von Rezessivität in Dominanz, nämlich von *j* in *J*.

Im folgenden werden zwei weitere Abzeichen der Testa hinsicht-
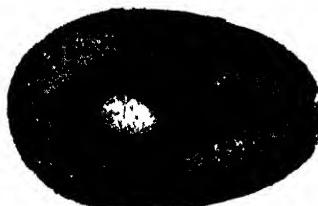
Fig 3.   Same der Testafarbe Veilchenartig Weiss mit hell Bister Hilumrand und Havannabrauner Bräme (Margo) um letzteren.

lich ihres Erbganges untersucht werden. Es sind dies die vom Hilumrand ausgehende Punktierung und die sog. Mikropylarpunkte.

DIE VERERBUNG DER PUNKTIERUNG DER TESTA.

Bei Punktierung beobachtet man auf der Samenschale zerstreut liegende Punkte in dunklerer, aber im übrigen ähnlicher Farbe wie die
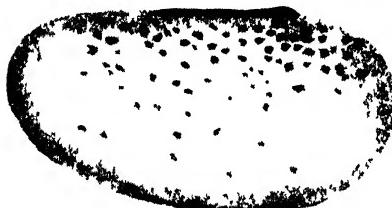
Fig 4.   Same mit punktierter Testa, Formel: *puncpunc*.

Testa. Ist z. B. die Testa Rhamninbraun, so sind die Punkte Dkl. Rhamninbraun bis Mineralbraun. Die Form der Punkte ist eine unregelmässige. Ihre Verteilung auf der Testa geht aus Fig. 4 hervor. Wie aus dieser ersichtlich, liegen die Punkte in der Nähe des Hilums dichter und auf der gegenüberliegenden Seite gewahrt man in der Regel nur ganz vereinzelte solche. Mitunter sind die Punkte überhaupt nur in der Nähe des Hilums wahrzunehmen. Bei okulärer Besichtigung machen diese Punkte den Eindruck von kleinen Grübchen. Eine nähere Untersuchung zeigt indessen, dass die Oberfläche

Fig. 5.   Querschnitt durch die Testa eines punktierten Samens. Die Punkte repräsentieren sich hier als dunkler gefärbte Zelleninseln in der Parenchymschicht.

keine Einsenkungen aufweist. Diese Erscheinung beruht zweifellos auf Lichtbrechung; denn ein Schnitt durch die Samenschale (vgl. Fig. 5)
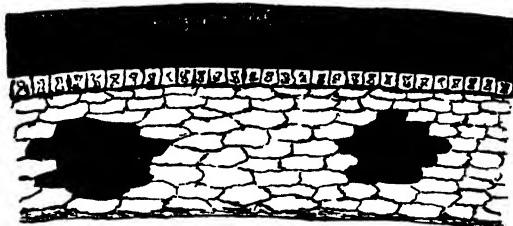
zeigt, dass diese Punkte auf farbige Zelleninseln in der tiefer gelegenen Parenchymschicht der Samenschale zurückzuführen sind.

Punktierte Samen wurden von mir zuerst auf einer Pflanze ange-troffen, die aus einer spontanen Kreuzung in der Sorte Flageolet Wachs stammte. Die Nachkommen dieser Pflanze waren konstant und wurden als Linie 157 weitergeführt. Die Testafarbe dieser Samen ist Rhamnin-braun. Mehrere Kreuzungen, ausgeführt zwischen dieser und nicht punktierten Linien zeigten, dass die Punktierung eine rezessive Eigen-schaft ist.

Kreuzung Nr. 118 wurde ausgeführt zwischen L. 157 und L. 27. Linie 27 stammt aus der französischen Wachsbohne De Digoin. Ihre Formel hinsichtlich Testafarbe ist $P\,c\,J\,g\,b\,v\,r$. Sie wurde mehrmals genetisch analysiert. F_1 dieser Kreuzung zeigte, wie zu erwarten, Rham-ninbraune Samen. Der Farbe Rhamninbraun kommt die Formel $P\,c\,J\,G\,B\,v\,r$ zu. Von einer Punktierung war auf diesen nichts wahr-zunehmen.

Die zweite Generation zeigte hinsichtlich Punktierung folgende Auf-spaltung: 570 ohne Punktierung : 196 mit Punktierung. Es handelte sich offenbar um eine monohybride Spaltung. D/m beträgt hierfür 0,38, also sehr gute Übereinstimmung mit den theoretisch erwarteten Spaltungszahlen anzeigend. Das für die Punktierung verantwortliche Gen belege ich, abgeleitet von der rezessiven Eigenschaft punktiert, mit dem Symbol *punc* (von *punctatus*).

Hinsichtlich Testafarbgenen fand noch Spaltung in $G : g$ und $B : b$ statt. In diesen beiden sowie im Genpaar *Punc—punc* ergaben sich zusammen folgende bifaktorielle Spaltungen.

Gefunden: 401 $G\,B$: 168 $G\,b$: 139 $g\,B$: 58 $g\,b$
Erwartet: 430,87   » : 143,62   » : 143,62   » : 47,87 »
D/m für
$9 : 3 : 3 : 1$ = — 2,17   + 2,26   — 0,43   + 1,51

Gefunden: 454 $G\,Punc$: 129 $G\,punc$: 116 $g\,Punc$: 67 $g\,punc$
Erwartet: 430,87   » : 143,62   » : 143,62   » : 47,87   »
D/m für
$9 : 3 : 3 : 1$.   + 1,68   — 1,35   — 2,56   + 2,86

Gefunden: 393 $B\,Punc$: 147 $B\,punc$: 177 $b\,Punc$: 49 $b\,punc$
Erwartet: 430,87   » : 143,62   » : 143,62   » : 47,87 »
D/m für
$9 : 3 : 3 : 1$ =   — 2,76   + 0,31   + 3,09   + 0,17

Da die letzte Spaltung, in den Genpaaren *B—b* und *Punc—punc,* grössere Abweichungen vom theoretisch erwarteten Verhältnis aufweist, werden noch die auf Grund der monohybriden Spaltungsverhältnisse zu erwartenden Werte mitgeteilt.

Erwartet: 402,1 *B Punc* : 137,9 *B punc* : 168,2 *b Punc* : 56,8 *b punc* .
D/m =   — 0,66   + 0,84   + 0,81   1,31

Nun besteht befriedigende Übereinstimmung. Die drei Gene *G*, *B* und *Punc* sind also wahrscheinlich nicht miteinander gekoppelt.

Mit Hinblick auf die durch das Gen *mar* bedingte lokale Umwandlung von Rezessivität in Dominanz des Gens *j* (vgl. Einleitung) entsteht hier analog die Frage, ob das Gen *punc* selbst für eine bestimmte Färbung der in Frage stehenden Zelleninseln in der Parenchymschicht der Testa verantwortlich ist, oder ob es dort nur die Wirkung gewisser Testafarbgene auslöst. Sicher festgestellt ist, dass die Farbe der Zelleninseln (mit Ausnahme von schwarzer Testa) stets eine dunklere ist als die den Pallisadenschicht. Im vorliegenden Fall ist die Pallisadenschicht Rhamninbraun, *P J G B*, die der Zelleninseln (= Punkte) am nächsten Mineralbraun, *P C J G B*. Es erscheint daher nicht unmöglich, dass das Gen *punc* in den Zelleninseln das Auftreten einer Farbe bedingt, die der genotypischen Konstitution der Testafarbe vermehrt durch die Wirkung von *C* entspricht. Dies würde bedeuten, dass bei Samen mit der Konstitution *C* die Farbe der Zelleninseln gleich der der Pallisaden sein sollte. Weitere Studien werden dies klarlegen.

DIE VERERBUNG DES ABZEICHENS MIKROPYLARPUNKTE.

Samen mit Mikropylarpunkten wurden von mir als Einmischung in einer in Ungarn vermehrten Partie von Flageolet Wachs gefunden. Sie zeigten die Testafarbe Blass Glaucescens, sollten demnach die genotypische Konstitution *P c j g b V r* besitzen. Dies konnte in Kreuzungsversuchen auch bestätigt werden. Im übrigen handelte es sich um eine ziemlich späte, niedrige Brechbohne.

Die Mikropylarpunkte bilden kleine unregelmässige Fleckchen in grünlich blaugrauer Farbe. Sie ist also sehr ähnlich der Testafarbe Blass Glaucescens, nur bedeutend dunkler. Eine anatomische Untersuchung der Testa ergibt, dass die Mikropylarpunkte aus eben so gefärbten Gruppen von Pallisadenzellen bestehen. Wie ihr Namen andeuten soll, nehmen sie ihren Ausgang von der Mikropyle oder deren unmittelbaren Nähe. Ihre Verbreitung auf der Testa ist am besten aus

Fig. 6 ersichtlich. Von der Mikropyle gehen bogenförmig seitlich nach unten divergierend zwei Gruppen von Punkten aus. Die Grösse dieser Punktegruppen variiert ziemlich stark in verschiedenen Linien. Bei gewissen können sich die Punkte über den grösseren Teil der Samenseiten erstrecken, bei anderen bilden sie nur je eine kleine Gruppe seitlich der Mikropyle (vgl. Fig. 6 unten).

Die nach der zuerst angetroffenen Pflanze mit Mikropylarpunkten gebaute Linie 98 zeigte die Punkte in mittlerer Verbreitung. Erst in Kreuzungen sind dann Typen mit verschiedener Ausbreitung erhalten worden Eine Kreuzung, Nr. 151, wurde ausgeführt zwischen L. 98 und L. 147. Letztere, eine Wachsbohne, stammt aus meiner Kreuzung Nr. 12 und hat die genotypische Konstitution $P c j g b v r$, sie ist demnach in allen Testafarbgenen rezessiv. Die auf F_1 dieser Kreuzung erhaltenen Samen zeigten die gleiche Testafarbe wie L. 98, also auch Mikropylarpunkte. Anscheinend stellten also die Mikropylarpunkte eine dominante Eigenschaft dar. Da die diesbezügliche genotypische Konstitution des anderen Elters, L 147, indessen nicht sicher bekannt war,

Fig 6   Zwei Samen mit dem Abzeichen Mikropylarpunkte auf der Testa Beim oberen Samen ist dieses Merkmal stark, beim unteren schwach ausgebildet

konnte erst F_2 hierauf endgültig antworten   Folgende Spaltung ergab sich.

Gefunden: 473 mit Mikropylarpunkten : 134 ohne Mikropylarpunkten
Erwartet: 455,25   »   : 151,75 »   »
D/m für 3 : 1 = 1,66.

Die Eigenschaft Mikropylarpunkte ist also dominant und zeigt monohybride Spaltung nach 3 : 1. Das hierfür verantwortliche Gen belege ich mit dem Symbol *Mip*, abgeleitet von der rezessiven Eigenschaft *Micropyle inpunctata* (nicht punktierte Mikropyle).

Linie 98 hat also die Formel $P c j g b V r Mip$, L. 147 $P c j g b v r mip$. Von Interesse ist nun, dass nicht nur Samen mit Testafarbe, im vorliegenden Fall Blass Glaucescens, sondern auch Reinweisse Samen ohne ein dominantes Testafarbgen, wohl aber mit dem Grund P, Mikro-

pylarpunkte hatten. Die Farbe dieser war dann nicht Grünlich Blaugrau sondern Blass Gelblich, etwa der Testafarbe Geschwefeltes Weiss nahe-kommend. Für die beiden Genpaare *V—v* und *Mip—mip* hat sich ergeben:

Gefunden: 358   *V Mip* : 90   *V mip* : 115   *v Mip* : 44   *v mip*
Erwartet: 341,44   »   : 113,81   »   : 113,81   »   : 37,94   »
D/m für
9 : 3 : 3 : 1 =   + 1,35   — 2,48   + 0,12   + 1,02

Die gefundenen Spaltungszahlen zeigen befriedigende Übereinstimmung mit den theoretisch erwarteten.

Weiter seien noch die Spaltungszahlen für das Genpaar *Y—y* (grüne—gelbe Hülsenfarbe) zusammen mit *Mip—mip* und *V—v* mit-geteilt.

Gefunden: 373   *Y Mip* : 95   *Y mip* : 100   *y Mip* : 39   *y mip*
Erwartet: 341,44   »   113,81   »   : 113,81   »   : 37,94   »
D/m für
9 : 3 : 3 : 1 =   + 2,58   — 1,96   - 1,44   ⊥ 0,18

Gefunden: 349   *Y V* : 119   *Y v* : 99   *y V* : 40   *y v*
Erwartet: 341,44   »   : 113,81   »   : 113,81   »   : 37,94   »
D/m für
9 : 3 : 3 : 1 =   + 0,62   + 0,54   — 1,54   + 0,35

Auch diese Zahlen zeigen befriedigende Übereinstimmung mit den erwarteten Verhältnissen, weshalb die Gene *Mip*, *Y* und *V* wahrschein-lich nicht miteinander gekoppelt sind.

Schliesslich sei erwähnt, dass auch die Mikropylarpunkte auf ver-schiedenen Testafarben in bestimmten, aber verschiedenen Farben auf-treten. Auch hier könnte hinsichtlich der Wirkung des Gens *Mip* Ana-loges gelten, was im vorigen Abschnitt für *punc* und früher für *mar* angeführt worden ist. Das Gen *Mip* könnte in den Punkten, in den diesen entsprechenden Gruppen von Pallisadenzellen, die Wirkung eines Farbgens (z. B. *C*) auslösen, d. h. dieses dort von Rezessivität in Do-minanz verwandeln, und so zusammen mit der übrigen genotypischen Konstitution für Testafarbe einen bestimmten Effekt hervorbringen. Hierbei muss auch auf eine eventuelle Beeinflussung der Farbenmodi-fikationsgene *Vir*, *Och* und *Flav* Rücksicht genommen werden. So könnte z. B. die grünlichblaugraue Farbe der Mikropylarpunkte von L. 98 der Formel *P C j g b V r Vir* entsprechen. Diese bedingt nämlich

Graugrüne Testafarbe. Auch hier werden natürlich erst Spezialstudien klaren Bescheid geben können.

ÜBER DIE WIRKUNG DER GENE V UND Inh.

Das Gen V ist seit langem bekannt (vgl. LAMPRECHT, 1932 a und 1935). Es sind drei Allelen bekannt, nämlich V, v_{lae} und v. Wahrscheinlich besteht noch ein viertes Allel v_{pal} (vgl. LAMPRECHT, 1936). Für die drei Allelen V, v_{lae} und v sind pleiotrope Effekte bekannt. Das Allel V bedingt Bischoffsviolette Blütenfarbe, Rote Stammfarbe und zusammen mit dem Grundgen P und Gri allein die Testafarbe Blass Glaucescens. Zusammen mit anderen Testafarbgenen verursacht V dunklere Farben in blauen—violetten Tönen bis Reinschwarz. Das Allel v_{lae} bedingt die Blütenfarbe Laeliafarbig und die Stammfarbe Rosa. Auf die Testafarbe scheint es keinen Einfluss zu besitzen. Bei Rezessivität in v sind die Blüten Weiss, der Stamm rein Grün und die Testa (bei Anwesenheit anderer Farbgene) Reinweiss.

Sehr charakteristisch für die Wirkung von V auf die Testafarben ist, dass die Wirkung dieses Gens beim Ausreifen und damit der Färbung der Samen stets zuletzt in Erscheinung tritt. Ein Beispiel möge dies illustrieren. Die Testafarbe Veilchenviolett hat die Formel $P C J V$. Der Formel $P C J v$ entspricht die Testafarbe Schamois. Beim Ausreifen der Samen zeigen diese nun stets zuerst die Farbe Schamois, die dann schmutzig Schamois wird, darauf hellvioletten Anflug bekommt und schliesslich dunkler bis zu Dunkel Veilchenviolett wird. Wenn solche Samen nicht gut ausreifen, so kann auch die endgültige Farbe ein schmutzig Braunviolett sein. Diese Umwandlung der Testafarbe geht stets vom Hilumrand aus und verbreitet sich allmählich über die ganze Samenschale. Am besten ist diese Farbe daher stets in der Nähe des Hilums ausgebildet. Genau das Gleiche gilt für alle Testafarben, die V in ihrer genotypischen Konstitution enthalten.

Worauf beruht nun dieser durch das Gen V bedingte, erst beim Ausreifen eintretende Ausfärbungsprozess? Die anatomische Untersuchung des Testaquerschnittes scheint hier Aufschluss gebenden Bescheid zu liefern. Es zeigt sich, dass die Pallisadenzellen bei allen Testafarben, ausgenommen mit V, gleichmässig gefärbt erscheinen. Bei Samen der Formel $P V$ enthalten diese Zellen einen grünlich blaugrauen Niederschlag in der Form von feinen Körnchen. Und dasselbe scheint für alle V in ihrer Formel enthaltenden Genotypen Gültigkeit zu haben. Ausgenommen hiervon sind natürlich sämtliche in den Grundgenen für

Testafarbe, *P* und *Gri*, Rezessiven, die überhaupt keinen Farbstoff aus-
bilden können.

Das Gen *V* verursacht demnach eine Fällungsreaktion, die erst bei
gewisser Konzentration des Zellsaftes während des Ausreifens einzu-
setzen beginnt. Dies erklärt auch, weshalb die Ausfärbung der Testa von
V-Samen in hohem Grade von den zur Zeit des Ausreifens herrschen-
den Umweltverhältnissen abhängig ist. Ausserdem dürfte diese Er-
scheinung auch von der übrigen genotypischen Konstitution beeinflusst
werden. So konnte ich schon lange beobachten, dass gewisse *V*-Linien
gewöhnlich typische dunkle Farbe zeigten, während andere niemals eine
solche erreichten, sondern meistens nur die entsprechende Farbe mit *v*
in rezessiver Form und etwas Anflug in der erwarteten Farbe hatten.
Besonders deutlich war dies hinsichtlich der Testafarben Graulich
Indigo, *P c J G B V*, Dkl. Indigo, *P c J g B V*, Ageratumblau, *P c J G b V*,
und Eisenhutviolett, *P c J g b V*.

Die vorhin erwähnte Kreuzung Nr. 151 scheint diesbezüglich Auf-
schluss zu geben. In dieser spalteten, abgesehen von *Y—y* für grüne—
gelbe Hülsenfarbe, die beiden Genpaare *V—v* und *Mip—mip*. Das Vor-
handensein von dominantem *V* konnte leicht stets an der Blütenfarbe
ermittelt werden. Mikropylarpunkte, *Mip* entsprechend, traten nun

			27 *Inh*	287	Blass Glaucescens mit Mikro-	
		36 *Mip*		256,08	pylarpunkten: *P V Mip Inh*.	
				(1,72)		
	48 *V*		9 *inh*	71	Reinweiss mit Mikropylar-	
				85,34	punkten: *P V Mip inh*.	
				(1,61)		
			9 *Inh*	74	Blass Glaucescens ohne Mik-	
		12 *mip*		85,34	ropylarpunkten: *P V mip Inh*.	
				(1,31)		
			3 *inh*	16	Reinweiss ohne Mikropylar-	
				28,45	punkte: *P V mip inh*.	
				(2,43)		
			9 *Inh*	115	Reinweiss mit Mikropylar-	
	16 *v*	12 *Mip*		113,81	punkten: *P v Mip (Inh inh)*.	
			3 *inh*	(0,12)		
			3 *Inh*	44	Reinweiss ohne Mikropylar-	
		4 *mip*		37,94	punkte: *P v mip (Inh inh)*.	
			1 *inh*	(1,02)		

F₁:
PP Vv Mip-
mip
Inhinh
Blass Glau- *F₂:*
cescens mit
Mikropylar-
punkten

Die *Aufspaltung des Bastards PP Vv Mipmip Inhinh in F₂ von Kreuzung Nr. 151.*
Vor den Farbenbezeichnungen steht die Anzahl gefundener Individuen, darunter die
erwartete Anzahl und in Klammern der Wert für D/m.

sowohl auf *V*- wie auf *v*-Samen auf. Im ersten Fall waren sie von grünlich-blaugrauer Farbe, im letzteren von hellgelber. Die *V*-Pflanzen konnten aber überdies noch in zwei Gruppen eingeteilt werden, eine mit der Testafarbe Blass Glaucescens und eine mit Reinweisser Testa. Beide hatten die Mikropylarpunkte in gleicher Farbe ausgebildet. Das beigegebene Spaltungsschema zeigt die hierfür erhaltenen Zahlen.

Wie ersichtlich wurde in dieser Kreuzung innerhalb der *V*-Pflanzen eine deutlich monohybride Spaltung nach 3 mit Blass Glaucescens : 1 Reinweiss gefunden. Für die letztere Gruppe ist also anzunehmen, dass die sonst beim Ausreifen der Samen durch *V* bedingte Ausfällung von grünlich-blaugrauen Körner im Zellsaft der Pallisadenzellen durch das Vorhandensein eines rezessiven Gens verhindert worden ist. Das hierfür verantwortliche Gen will ich, abgeleitet von *inhibeo* = hemmen, mit dem Symbol *Inh* bezeichnen. Eine sichere Klassifikation in bezug auf dieses Gen wird allerdings nicht stets möglich sein. Nur in Kreuzungen mit einigermassen gleichzeitigem Ausreifen werden mit dem theoretisch erwarteten Verhältnis nahe übereinstimmende Werte erhältlich sein. Bei auch hinsichtlich Reifezeit stärker spaltenden Kreuzungen wird die Spaltung im Genpaar *Inh—inh* durch den ähnlich wirkenden Einfluss der Umweltverhältnisse die Grenzen verwischen. — Zu ermitteln verbleibt ob *inh* bei allen Testafarben mit *V* die Wirkung dieses Gens vollständig inhibiert oder nur stark abschwächt. Letzteres wurde wiederholt beobachtet, ohne dass jedoch genetische Analysen ausgeführt worden sind.

SUMMARY.

1. The inheritance of two new points of different colour on the seed coat of *Phaseolus vulgaris* is studied.

2. One of these, the character dotting, is caused by coloured islands of cells in the parenchymal layer of the seed coat (Figs. 4 and 5). This character is singly recessive and corresponds to the new gene pair *Punc—punc*.

3. The second character is the so-called micropyle points. The development of these coloured points always begin in the neighbourhood of the micropyle (see Fig. 6). Micropyle points are a dominant character, which correspond to the new gene pair *Mip—mip* (derived from *micropyle inpunctata*).

4. The effect of the gene *V*, causing the seed coat colour Pale Glaucescens, has been closely studied. It may be mentioned that this

gene causes the precipitation of dark glaucescent fine grains in the pallisade cells, which form the epidermis.

5. A new gene *Inh* (derived from *inhibeo*) has been found, which in its recessive condition, *inh*, prevents the precipitation of glaucescent grains, caused by *V*.

XVIII. ÜBER MATTE SAMENSCHALE UND IHRE VERERBUNG.

Nicht alle Farben der Samen von *Phaseolus vulgaris* zeigen gleich hohen Glanz.   So sind z. B. Farben ohne das dominante Gen *J* gewöhn-
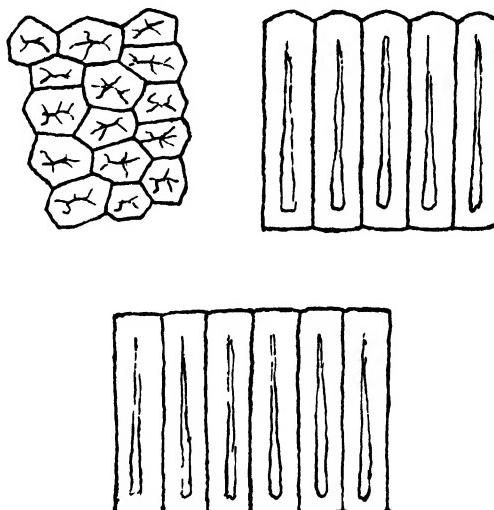
Fig. 7. Oben, links: Flächenansicht der Pallisadenzellen der matten Testa (*aspasp*), rechts: Längsschnitt durch dieselben. Man beachte die Kuppenbildung am oberen (äusseren) Ende. Unten: Längsschnitt durch Pallisadenzellen der normalen, glänzenden Testa (ohne Kuppenbildung).

lich etwas bis deutlich weniger glänzend als solche mit *J*. Stark glänzend ist z. B. Rohseidengelb, *P J*, Schamois, *P C J*, Bister, *P C J G* usw. Weniger stark glänzend bis matt sind Veilchenartig Weiss, *P B*, Ambraweiss, *P C B* usw. Die Struktur der äussersten Zellenschicht solcher mehr oder weniger glänzender Typen zeigt indessen kaum welche sicheren Unterschiede. Die Mattigkeit des im folgenden zu besprechenden Typus ist von diesen stark verschieden, sowohl bei okulärer Betrachtung wie bei anatomischer Untersuchung.

Stark matte Samen wurden von mir aus dem Keniagebiet in Afrika erhalten. Eine Linie dieser mit schwarzer Testafarbe wurde unter Nr. 61

weitergebaut. Die Oberfläche der Samen von L. 61 erscheint, typisch sammtartig matt. Bei etwa 20-facher Lupenvergrösserung erscheint die Oberfläche deutlich chagriniert, also körnig. Ein Schnitt durch die Testa gibt diesbezüglich Bescheid und zeigt einen markanten Unterschied gegenüber dem normalen glatten Typus. Wie **Fig. 7 zeigt,** sind die äusseren Enden der Pallisadenzellen durch eine Kuppe abgeschlossen. Hierdurch wird sowohl die Körnelung der Oberfläche bedingt wie auch im Zusammenhang mit der dadurch verursachten ungleichen Reflexion des Lichtes das sammtartig matte Aussehen. Auch sind die Risse auf den Kuppen stärker zutagetretend als auf den sonst flachen Zellenenden.

Mit Linie 61 wurden mehrere Kreuzungen ausgeführt. Zu Kr. Nr. 148 wurde als zweiter Elter Linie 147 verwendet, die nur in den beiden Grundgenen *P* und *Gri* dominant, in allen Testafarbgenen aber rezessiv ist. Die auf F_1 erhaltenen Samen zeigten glatte Oberfläche, die Mattigkeit, die Rauheit der Testa scheint demnach ein rezessives Merkmal darzustellen. In der zweiten Generation wurden insgesamt 943 Pflanzen untersucht, die folgendermassen spalteten:

Gefunden: 672   mit glatter Testa : 271   mit matter Testa
Erwartet: 707,25   »   »   » : 235,75   »   »   »
D/m für 3 : 1 = 2,65.

Die Übereinstimmung zwischen gefundenen und bei Monohybridie theoretisch erwartetem Spaltungsverhältnis ist keine gerade gute, aber zweifellos noch als monohybrid aufzufassen. Das für die Spaltung in glatte : matte (rauhe) Testa verantwortliche Genpaar belege ich mit den Symbolen *Asp—asp*, abgeleitet von der rezessiven Eigenschaft *asper =* rauh.

SUMMARY.

1. Seeds of *Phaseolus vulgaris* with a rough, lustreless surface were received from Kenya, Africa.

2. The roughness of the seed coat is caused by elevations (like cupolas) of the ends of the pallisade cells, which form the epidermis (Fig. 1).

3. The character of roughness of the seed coat is singly recessive, corresponding to the gene pair *Asp—asp* (derived from *asper =* rough).

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7. — 1939. Lokale Umwandlung von Rezessivität in Dominanz durch die Wirkung eines besonderen Gens bei *Phaseolus vulgaris*. — Z. f. ind. Abst. u. Vererbgl. LXXVII, 186—194.

INHALT.

CYTOLOGY OF AGROPYRON JUNCEUM, A. REPENS AND THEIR SPONTANEOUS HYBRIDS

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THE material for this study was obtained from natural localities in South Sweden. The root tips and anthers were fixed in chrome-acetic-formalin. The preparations were stained with gentian violet.

Somatic cytology. — *Agropyron junceum* (L.) P. B. was found to have 2n = 28. Material from five localities was studied. Counts were made on 9 plants, 7 determinations were exact (3 plates), and 2 approximate.

The chromosome number 2n = 28 was previously observed in this species by PETO (1930), SIMONET (1934, 1935 a and b), PARDI (1937), and SIMONET and GUINOCHET (1938). It was found that *A. junceum* consists of two subspecies, an atlantic form with 2n = 28, and a Mediterranean form with 2n = 42 (SIMONET, 1935 a and b; PARDI, 1937; SIMONET and GUINOCHET, 1938).

SIMONET (1935 b) observed two secondary constrictions in the somatic chromosome complement of the tetraploid type. In 1934 he observed none, his material was then from another locality. PARDI (1937) observed none.

The present writer observed no secondary constrictions in most plates, but in some one such constriction was observed, and in a few two. The observations differed in this respect even between cells belonging to the same plant.

It seems probable to me that the contradictory statements as to the number of secondary constrictions are due to the use of inferior fixatives, and that their actual number may be greater than ever observed. It was pointed out by FLOVIK (1938) that chrome-acetic-formalin fixatives are poorly suited for the study of chromosome morphology, although they are good for the determination of chromosome numbers (l. c. pp. 270—271, 278, and Figs. 6—7).

As pointed out by HEITZ (1931), there is a close relation between secondary constrictions and nucleoli. Although I am not convinced

that all secondary constrictions must be nucleolar organizers, and that all nucleoli are organized by constrictions, it is of interest in this connexion to determine the maximum number of nucleoli. In numerous nuclei in the tetrad cells two nucleoli were found. At interphase four nucleoli were clearly observed in a few cells. As the chromosomes are double at interphase it is not certain that this number really proves the existence of four nucleolar organizers in the gametic chromosome set. That there are at least two is rather certain, however.

Agropyron repens (L.) P. B. (4 clones from 3 localities) was found to have 2n = 42. The determinations are approximate. There are no reasons, however, to believe that the plants are aneuploids.

This chromosome number has been reported also by STOLZE (1925), MOWERY (1929), PETO (1929 and 1930), AVDULOW (1931), SIMONET (1935 a) and NIELSEN and HUMPHREY (1937). PETO (1930) found 2n = 35 in material from U.S.S.R. AVDULOW found 2n = 28 in a material which probably is also from U.S.S.R., as this author is a Russian.

Hybrids between *A. junceum* and *A. repens* from five Swedish localities were found to have 2n = 35. As there are apparent morphological differences between some plants collected in the same locality, the number of different clones with this number is 7 (4 determined exactly and 3 approximately). A number of plants from Falsterbo (province of Skåne, South Sweden) had 2n = 49. As they are morphologically very much alike, they no doubt belong to the same clone. All observations did not give exactly 49 chromosomes, but the variation may be ascribed to the difficulties of making exact counts. These hybrids are then pentaploid and heptaploid, as the basic number of the genus is 7.

35 is the chromosome number to be expected in a hybrid between parents with 28 and 42 chromosomes. This chromosome number was found in the similar hybrids, *A. junceum* (L.) P. B. × *A. littoreum* (SCH.) ROUY (SIMONET, 1934) and *A. junceum* (L.) P. B. × *A. littorale* (HOST) DUM. (SIMONET, 1935 a). The hybrid with 2n = 49 is supposed to contain two gametic chromosome sets from *A. junceum* and one from *A. repens* (14 + 14 + 21). (Origin discussed below.) This chromosome number has been observed previously in *Agropyron* in the hybrid between *A. repens* (L.) P. B. (2n = 42) and *A. campestre* G. G. (2n = 56) (SIMONET, 1935 a).

The chromosomes of *A. junceum* seem to be largest, those of *A. repens* smallest, and those of the hybrids intermediate (Figs. 1—4

and 5—6). It might be suggested that a higher chromosome number causes a smaller chromosome size. Evidence against such an assumption is the fact that PARDI (1937) has found the chromosome size of the tetraploid and the hexaploid types of *A. junceum* to be the same (illustrated with drawings and photographs). The differences in my
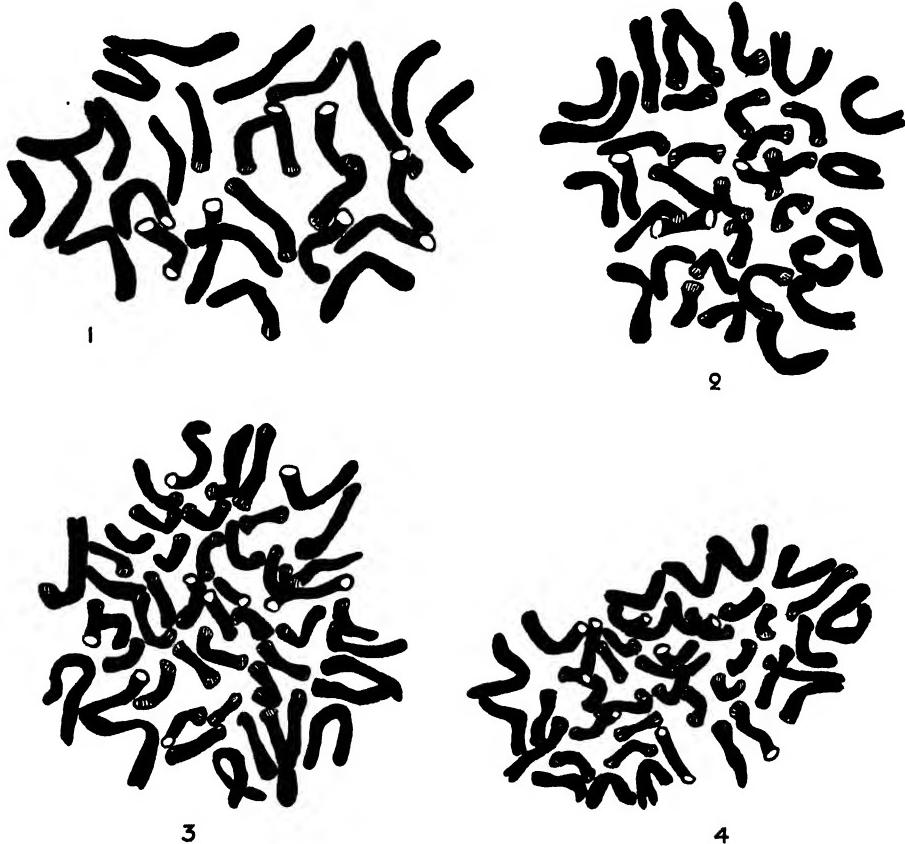
Figs. 1—4. Somatic metaphases. — Fig. 1. *Agropyron junceum,* 2n = 28; Fig. 2, *A. junceum* × *repens,* 2n = 35; Fig. 3, *A. junceum* × *repens,* 2n = 49; Fig. 4, *A. repens,* 2n = 42. — × 2500.

case are probably due to genotypic differences between the two species. (It is known that chromosome size may be genotypically controlled, e. g. DARLINGTON, 1937, pp. 52—60.)

Fertility. — 8 individuals of *A. junceum* were studied as to pollen fertility. 4 of them had a percentage of good pollen of 90—100, 3 had 80—90, and one 70—80. The one with 70—80 % had 107 bad grains in a total of 388 grains. Only one pollen test was made on each indi-

vidual. The tests were made with free pollen and not by crushing the anthers. 3 individuals of *A. repens* were found to have 90—100 % good pollen.

Seed fertility in *A junceum* was studied on spikes gathered from a natural population. The two lowermost flowers in each spikelet were studied. 50 flowers were sterile in a total of 147. RAUNKIAER (1927, p. 344) found 54 % sterility in two hundred spikes of *A. repens*.

Three of the pentaploid hybrids were studied as to pollen fertility. In more than 4000 pollen grains of these plants 3 morphologically good grains were found. The others were quite empty and shrivelled. In the heptaploid hybrid 5 good grains were found in more than 1000. Similar results were obtained in other preparations of this hybrid, and in a preparation made of it in the preceding summer (1938) (grains not counted). The anthers of the hybrids do not dehisce.

No seeds were found in 388 spikelets of the pentaploid hybrids, and in 1496 spikelets of the heptaploid hybrid, after open flowering.

Meiosis in the pure species. — First metaphase in the pure species gives, when regular, 14_{II} in *A. junceum* and 21_{II} in *A. repens* (Figs. 5—6). I have seen no previous statements as to meiosis in *A. junceum*. 21_{II} were found in *A repens* by STOLZE (1925), MOWERY (1929), and PETO (1930).

I also found some irregularities in the two species. Four individuals of *A. junceum* were studied, and all of them were found to be heterozygous for at least one inversion. This was shown by the simultaneous occurrence at first anaphase of a chromosome bridge and a fragment (Fig. 10). In some cases bridges were observed which were not accompanied by fragments. They may have been caused by a delayed chiasma separation. The frequency of inversion bridges is rather low, being estimated at 1—2 per cent approximately.

Univalents were observed in all the four individuals. At metaphase they are distributed at random in the cell or lying at the periphery of the plate (Fig. 8). At anaphase they often lag and divide (Fig. 9). The frequency of cells with univalents was estimated at 10 per cent approximately.

Quadrivalents were observed in three of the individuals. The reason why no quadrivalents were found in the fourth individual may be due to the fact that the material studied of this individual was rather small, or it may be due to constitutional differences.

Seven individuals of *A. repens* were studied. Inversion bridges at first anaphase were found in none, but the material studied of this
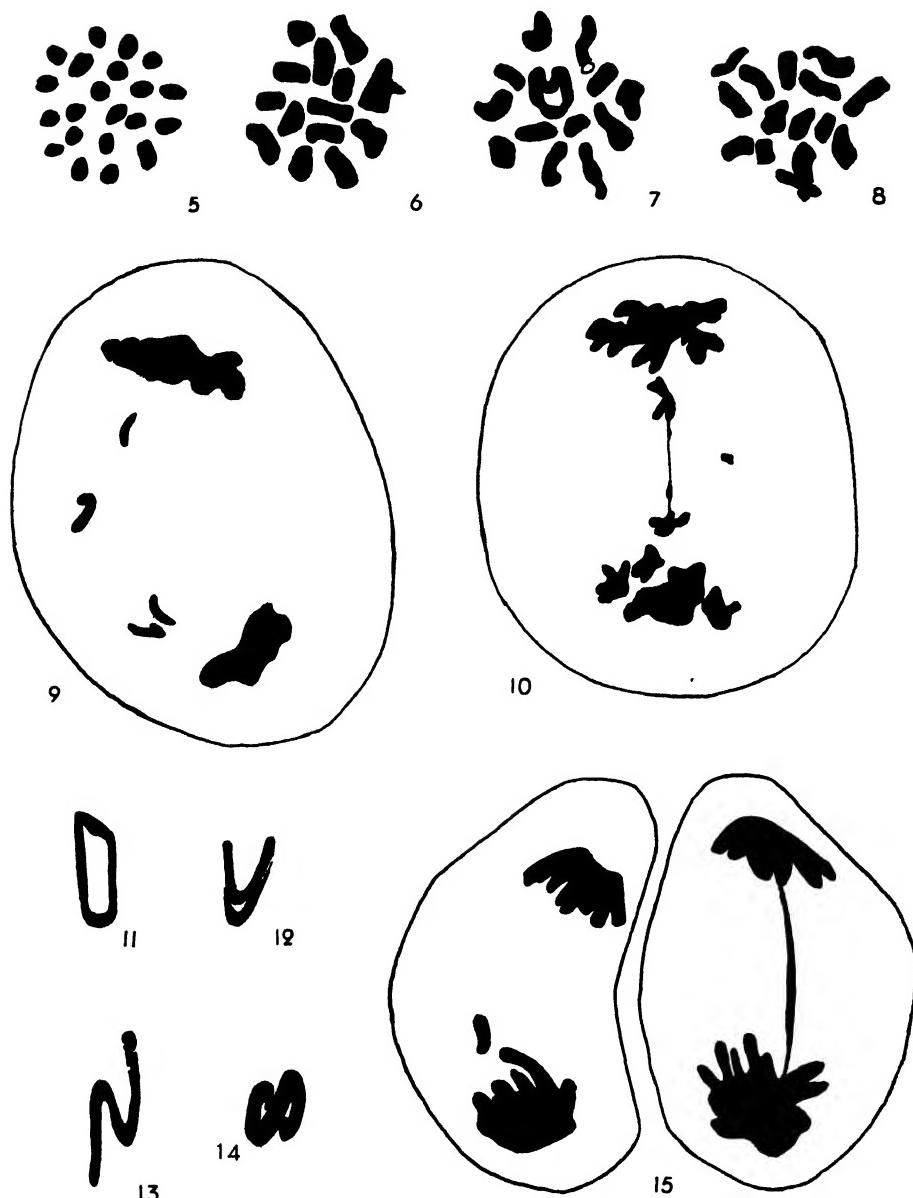
Figs. 1—15. Meiosis in the pure species. — Figs. 5—8, first metaphases; Fig. 5, *A. repens*, 21_{II}; Figs. 6—8, *A. junceum*; Fig. 6, 14_{II}; Fig. 7, $12_{II} + 1_{IV}$; Fig. 8, $13_{II} + 2_{I}$; Figs. 9—14, *A. junceum*; Figs. 9—10, first anaphase; Fig. 9, two dividing univalents; Fig. 10, a chromosome bridge and a fragment; Figs. 11—14, quadrivalents drawn from different first metaphases; Fig. 15, *A. repens*, second anaphase with chromosome bridge. — $\times 2200$.

stage was rather small. In acetocarmine preparations of two indi-
viduals second anaphase bridges were observed. At the second division
a bridge can scarcely be due to a delayed chiasma separation, and I can
imagine no other cause than a dicentric chromatid, which may be the
result of dyscentric hybridity. These bridges were not, however, ac-
companied by clear fragments.

Univalents were present in all the *repens* plants studied. Quadri-
valents were plainly observed in two individuals, and probably in four
others.

It is not possible from these observations of quadrivalents in the
two species to state with certainty whether they are caused by seg-
mental interchange or autopolyploidy, which may be partial or com-
plete. I feel inclined, however, to ascribe their occurrence to structural
hybridity.

Nor can it be said with certainty that the inversion bridges are due
to structural hybridity in the usual sense. They may arise from
occasional pairing between chromosomes belonging to different
genomes in an allopolyploid, if these genomes differ by inversions.
The univalents also may indicate structural hybridity, but that, too, is
not certain.

Univalents and quadrivalents were found in the hexaploid species
A. glaucum by PETO (1936). The quadrivalents in this species were
supposed to be caused by segmental interchange (l. c. p. 205). In
A. elongatum (2n = 70) he observed many higher configurations, even
octavalents.

If *A. junceum* and *A. repens* are cross-fertilisers, which is probable,
judging from their open flowering, the meiotic irregularities found in
them might be connected with this mode of reproduction. Structural
hybridity seems to be rather common in cross-fertilising plants
(MÜNTZING, 1939).

Another fact which might have some influence in this relation is
the pronounced vegetative propagation of the species in question. As
pointed out by DARLINGTON (1937, p. 271), the hybridity equilibrium in
regard to inversions is highest in plants that are largely propagated by
asexual means.

For further details on structural hybridity in species and hybrids,
see DARLINGTON (1937) and MÜNTZING (1939).

Meiosis in the hybrids. — Meiosis in four pentaploid hybrids was
studied. The following frequencies of bivalents and univalents were
found.

| Univalents	Bivalents	Number of cells
9	13	3
11	12	4
13	11	5
17	9	1
Average 11,8	11,6	Total 13

Five cells were studied in one individual, and four cells in each of two others. Judging from the appearance of numerous other cells, the amount of pairing was about the same in them. This is also true of the fourth individual, in which no cell was analysed. In aceto-carmine preparations some possible trivalents were seen. — In one cell in an acetocarmine preparation there seemed to be 35 univalents distributed at random in the cell. The neighbouring cells all showed an amount of pairing of the same order as that recorded in the table.

At first anaphase there often occurs chromosome bridges and fragments, and even more than one bridge in the same cell (Fig. 17). Undoubtedly this hybrid is heterozygous for some inversions. There may have occurred more bridges in one of the individuals than in the others, but I cannot say if the difference is certain. Bridges and fragments were observed at the second division too. Many univalents often lagged and divided at first anaphase. In Fig. 18 there are three cells at second division. The small cell has probably been formed by some univalents. This figure also shows lagging univalents. The tetrad cells often contained extra nuclei and chromatin fragments (Fig. 19). These are probably formed by univalents and acentric fragments.

Thus, it is evident that the hybrid studied is not only a numerical hybrid, but also a structural one. The structural hybridity is indicated also by a reduced chiasma frequency in the pairing chromosomes. It could be seen at once that the proportion of rod-shaped bivalents is much higher in the hybrid than in its parents. The simultaneous occurrence of structural and numerical hybridity is probably the reason why the hybrid is so extremely sterile.

Although the amount of pairing does not deviate very much from that expected on the assumption that the two species have two genomes in common, such a thing cannot, of course, be considered established by the present observations. The pairing might also be due chiefly to autosyndesis, or all the five genomes may be more or less homologous to each other. Assuming, however, that the genotype of the hybrid does not cause a reduced pairing (e. g. by special genes reducing

chiasma frequency), one conclusion can be drawn from the pairing found, viz. that both the species are not completely autopolyploid. There are also other reasons indicating that *A. junceum* is not completely autopolyploid (ÖSTERGREN, 1940 b).
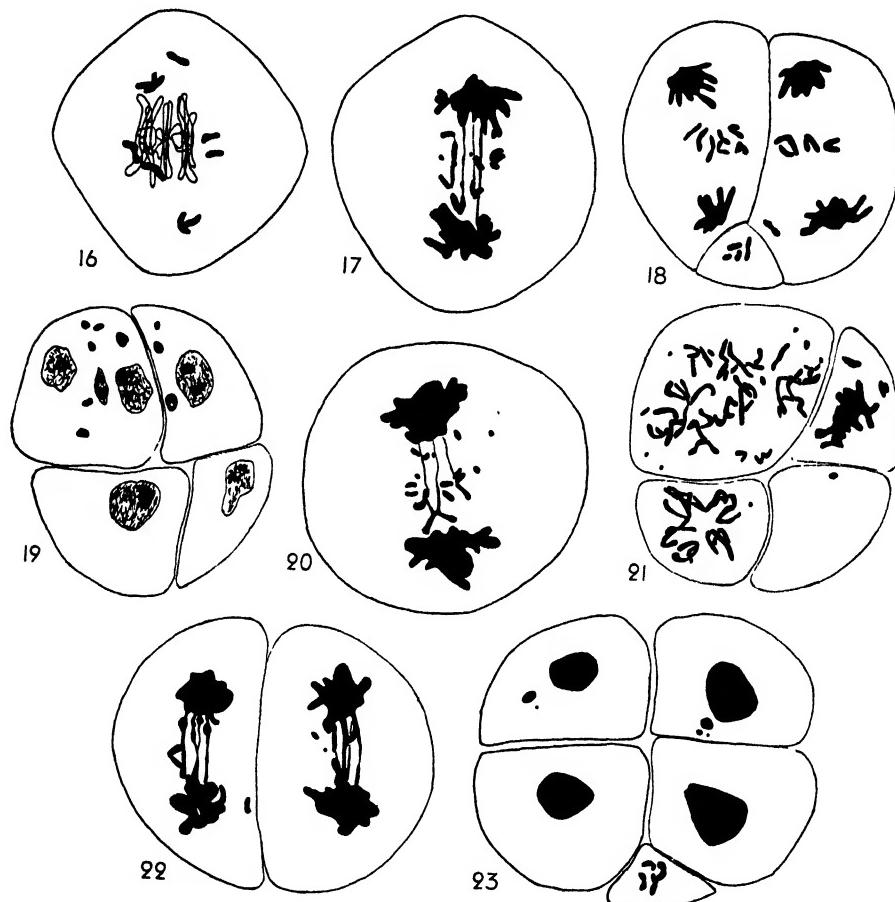
Figs. 16—23. Meiosis in the hybrids. — Figs. 16—19, the pentaploid hybrid; Fig. 16, first metaphase, $13_{II} + 9_I$; Fig. 17, first anaphase with bridges; Fig. 18, second anaphase with lagging univalents, and an extra cell with a few chromosomes; Fig. 19, tetrad. — Figs. 20—23, the heptaploid hybrid; Fig. 20, first anaphase, see text; Fig. 21, a supposed second metaphase after a multipolar first division spindle; Fig. 22, second anaphase with chromatin bridges; Fig. 23, tetrad with an extra cell. — × 1400.

According to DARLINGTON (1937, p. 218), the pairing in the hybrid studied by SIMONET (1934) is presumably similar to that of pentaploid *Triticum* and *Nicotiana*, four chromosome sets being associated as pairs

and one remaining chiefly unpaired. The amount of pairing should then not differ very much from that found in my pentaploid hybrids. Nothing is mentioned, however, in SIMONET's paper (1934) as to meiosis in his hybrid, and I do not know the reasons for DARLINGTON's presumption.

If we ascribe *A. junceum* and *A. repens* the genome formulae $J_1J_1J_2J_2$ and $R_1R_1R_2R_2R_3R_3$, the formula of the heptaploid hybrid will be $J_1J_1J_2J_2R_1R_2R_3$, according to the assumptions as to its constitution made above. If *A. repens* were a pronounced allopolyploid, and its chromosomes were not homologous with those of *A. junceum*, the three chromosome sets from *repens* would appear as 21 univalents. If *A. repens* is partly autopolyploid or its chromosomes more or less homologous to those of *A. junceum*, the number of univalents will be reduced by autosyndesis and the formation of trivalents (or other multivalents) with the chromosomes of *A. junceum*.

Because of the high chromosome number of the heptaploid hybrid no first metaphase configurations could be completely analysed. The number of univalents lying apart from the equatorial plate was counted in 77 cells and an average of 4,61 was found. The lowest number, 0, was found in 7 cases and the highest, 12, in one case. In 28 first anaphase cells the number of lagging univalents gave an average of 3,93. It is evident that these numbers must be lower than the actual number of univalents. It does not seem very probable, however, that it amounts to 21. Thus it may be safely concluded that one or both of the factors mentioned above is at work.

The number of chromatin bridges at first anaphase varied in 28 cells from 0 to 3, with an average of 1,13 per cell. The number of fragments varied in these cells from 0 to 7, with an average of 1,68. Fig. 20 shows an interesting case. There is one bridge of the common type, and two others, and these two are both attached to the same centromere. This centromere also carries two normal chromatid arms, as expected under such circumstances. This configuration has probably arisen from dyscentric hybridity in a trivalent. It may also have arisen from a bivalent, but then we should have expected the two bridges to lie more close together. This figure shows dividing univalents and fragments too.

Chromatin fragments are common also at interphase, second metaphase, second anaphase and telophase and in the tetrads. At second anaphase, too, there are often chromatin bridges. The number of cells resulting from the first division is in most cases two, but some excep-

tions were found. Fig. 21 is interpreted as a second division after a multipolar first division spindle. In the lower left cell there are 6 chromosomes. The lower right cell is supposed to have been damaged.

The number of cells in the tetrads is usually 4, but a few cases with 5 were found. In Fig. 23 there are four chromosomes in a supposed anaphase position in the small cell, two in an upper level and two in a lower one. This extra cell has probably been formed by two univalents.

Discussion. — If the heptaploid hybrid contains two gametic chromosome sets from *A. junceum* and one from *A. repens*, as assumed above, it should be expected to resemble *A. junceum* more closely in morphology than the pentaploid hybrids. This is also the case with respect to many characters (ÖSTERGREN, 1940 a; it has for certain reasons been considered appropriate to treat the morphology of these plants in a special paper).

There are two ways in which this heptaploid hybrid may have arisen. The first is the production of an unreduced gamete in a pentaploid hybrid and the fertilisation of this gamete with a normal one from *A. junceum.* There are numerous cases known of non-reduction in hybrids (see, e. g., the table by DARLINGTON, 1937, p. 417). The second possibility is the production of an unreduced gamete in the pure species *A. junceum* and the fertilisation of this with a normal one from *A. repens.* Unreduced gametes in pure species have been found in crosses between *Phleum* species by NORDENSKIÖLD (1937). In a certain cross it was even found that the only gametes functioning on the female side were the unreduced and the doubly unreduced ones (l. c. pp. 306— 309). Then the chromosome numbers of the gametes functioning were more alike than they would have been if the reduced gametes had functioned. MÜNTZING (1936) considers such cases to be due to the viability of a zygote being greater the more alike the chromosome numbers are of the gametes giving rise to it. He also gives some other examples (l. c. pp. 326—334). Such a thing might have some influence in my case, too, as the difference between 28 and 21 is relatively smaller than that between 21 and 14.

If *A. junceum* can produce unreduced gametes, this fact might be of interest in the discussion of the origin of the hexaploid subspecies found by SIMONET (1935 a and b). If such an unreduced gamete was fertilised by a normal one, the result would be just a hexaploid individual. It is suggested by SIMONET and GUINOCHET (1938) that the hexaploid type might in some way have arisen from the tetraploid one. I believe, however, that the tetraploid type is an allopolyploid rather

than an autopolyploid. In such a case it could not at once give rise to a fertile and constant hexaploid race, but such a race might, of course, arise in later generations.

The discovery that the hybrid between *A. junceum* and *A. repens* consists of two caryologically different types contributes to the explanation of the variation for which this hybrid is well-known. The variation is, however, caused by other circumstances as well (see ÖSTERGREN, 1940 a).

SUMMARY.

Agropyron junceum has 2n = 28, and *Agropyron repens* 2n = 42. as found also by previous workers. Seven spontaneous hybrids (from South Sweden) between *A. junceum* and *A. repens* were found to have 2n = 35, one hybrid had 2n = 49. The hybrids are highly sterile. Meiosis in the pure species was not always regular, inversion bridges, univalents, and quadrivalents were found. 13 first metaphases in the pentaploid hybrids gave an average of $11{,}6_{II} + 11{,}8_{I}$. In the heptaploid hybrid the chromosomes of *A. repens* are supposed to pair autosyndetically or as trivalents with the *junceum*-chromosomes, to some extent. Both hybrids are heterozygous for inversions. It is assumed that the heptaploid hybrid has arisen from the back-cross of a pentaploid hybrid with *A. junceum* or from the fertilisation of an unreduced gamete of *A. junceum* with a normal one from *A. repens*.

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NOTE ON THE SOMATIC CHROMOSOMES OF SOME COLCHICUM SPECIES

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DUE to their immunity to colchicine and sensitivity to other chemicals with a similar action as colchicine (LEVAN, 1940), the chromosomes of *Colchicum* must be considered to be of special cytological interest. Since extremely few cytological data are published on this genus, I shall in the present paper give a short account of a few observations made in connection with chromosome-physiological studies in *Colchicum*.

Colchicum has proved to be a rather difficult cytological material. FURLANI (1904) gives the chromosome number of *Colchicum autumnale* as n = 7, and HEIMANN-WINAWER (1919) gives the number n = 10—12 for the same species. Both these numbers are probably erroneous. MILLER (1930) writes: »Although 5 species of *Colchicum* have been examined, no definite number can be given for any of them. The chromosomes would appear to be extremely adhesive, and hang together in chains from 2 to 5 . . . The diploid number so far obtained for *Colchicum* is certainly more than 24, probably more than 32, and quite possibly more than 40 . . . The adhesiveness seen in the chromosomes of the group *Colchicae* is present also in some of the other groups (of the *Melanthioideae* section), although not to such an extent». NEWTON (in TISCHLER, 1931) gives n = 21 for the two species *Colchicum autumnale* and *Parkinsonii*. SUTÔ in his list of Liliaceous chromosomes (1936) mentions *Colchicum* as having the *Lilium-Narcissus* type of its idiogram, i. e. all the chromosomes large and of about the same length. I have not been able to find from where this erroneous description originates, since, as far as I am aware, no pictures or detailed descriptions of *Colchicum* chromosomes have so far been published.

In the present paper 10 *Colchicum* species are examined (Table 1). They were procured from the Botanical Gardens of Lund and Copenhagen by the courtesy of Mr. A. TÖRJE and Dr. O. HAGERUP. All of them belong to the Sectio *Autumnales* (STEFANOFF, 1926), with the exception of *montanum*, the determination of which is not sure. The root tips were fixed in NAVASHIN. In order to diminish the tendency

of adhesiveness of the chromosomes, the fixations were made under cold conditions (at about 0° C), and the plants were kept in a cold greenhouse. The chromosomes were stained in gentian violet. The plants studied showed an abundancy of beautiful mitoses. Nevertheless there was a risk of making erroneous estimations of the chromosome numbers, since the large chromosomes very often had prominent
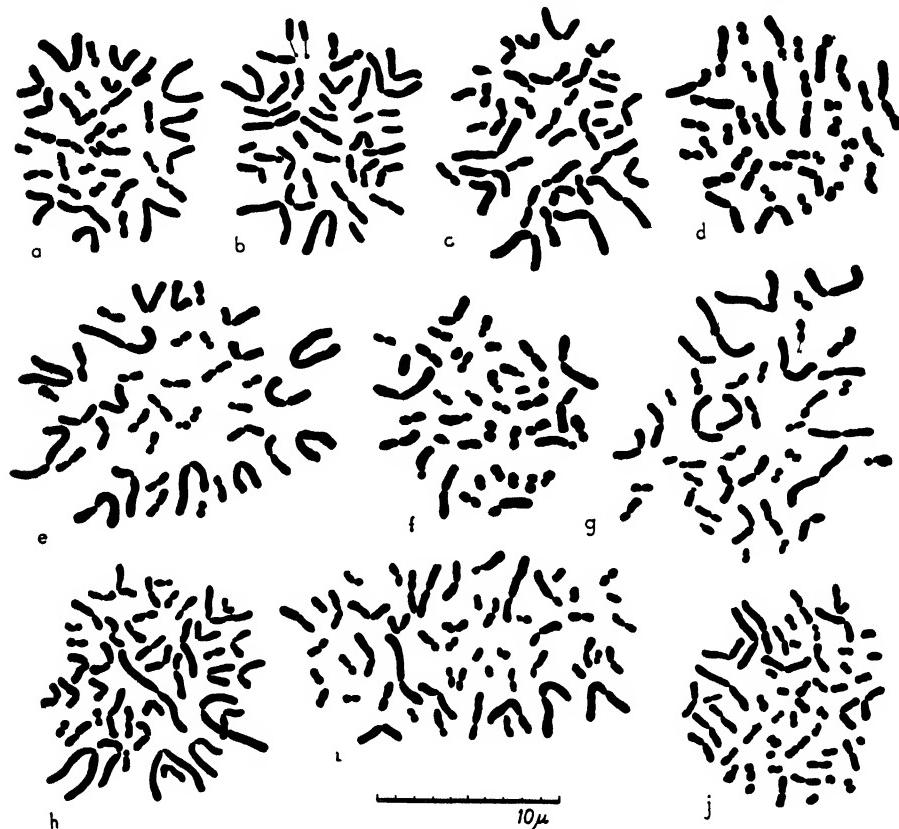

Fig. 1.   Somatic metaphase plates of *Colchicum* species, *a*: *Bivonae* (2n = 36), *b*: *autumnale* (2n = 38), *c*: *neapolitanum* (2n = 38), *d*: *speciosum* (2n = 38), *e*: *byzantinum* (2n = 40), *f*: *giganteum* (2n = 40), *g*: *Bornmülleri* (2n = 42), *h*: *variegatum* (2n = 44), *i*: *latifolium* (2n = 54), *j*: *montanum* (2n = 54). — × 2400.

constrictions, due to which a chromosome segment could easily be interpreted as a free, small chromosome. The chromosome numbers given in Table 1 are probably correct, however, although in some forms of *speciosum* 39 were counted instead of 38 chromosomes, and in some plants of *autumnale* the numbers 39 and 40 were counted. Possibly a certain degree of autopolyploidy is present, which, as for instance in

Allium, might cause the origin of clones with a somewhat varying somatic number. The examination of meiosis should settle this question. It is certain, however, that the differences of the idiograms between the species is not only of a numerical nature, but also a structural. In most species, for instance, the largest chromosomes have medially located centromeres, while in some species (e. g. *giganteum*) the largest chromosomes are clearly asymmetric.

The appearance of the chromosomes of the different species is seen from Fig. 1. The variation in size within each idiogram is very prominent. 4—6 chromosomes of each species are very long, up to 7 μ. These chromosomes have ordinarily median centromeric constriction. In certain species 2 of these chromosomes may be recognized by the

TABLE 1. *The Colchicum species examined.*

Species	2n	Pictured in Fig. 1	Material procured from
Bivonae GUSS.	36	1a	Copenhagen
autumnale L.	38	1b	Lund, Copenhagen
neapolitanum TEN.	38	1c	Copenhagen
speciosum STEV.	38	1d	Lund, Copenhagen
byzantinum TEN.	40	1e	Copenhagen
giganteum hort.	40	1f	Copenhagen
Bornmülleri FREYN.	42	1g	Lund
variegatum L.	44	1h	Copenhagen
latifolium S. S.	54	1i	Copenhagen
montanum L.	54	1j	Copenhagen

presence of a secondary constriction in about the middle of one arm (Fig. 1 *d*). A great many chromosomes of medium size are present. Their size varies continuously, and I have not made any attempt to classify them. Most of them are more or less medially inserted, but in most species some subterminally inserted chromosomes of the medium size-class occur. In a few cases the shorter arm of these chromosomes is furnished with a satellite (Fig. 1 *d*). Among the small chromosomes there exist differences in size, the smallest are not longer than 0,6—0,8 μ, thus only about one tenth of the longest chromosomes. Also among the small chromosomes the medially attached types predominate, even if purely terminally attached chromosomes seem to occur at least among the smallest chromosomes, which are almost spherical at metaphase. In a couple of species one pair of the small

chromosomes has satellites (Fig. 1 b, g). The satellites are tiny and the attachment threads are long.

The examples of idiograms of *Colchicum* species given above show that several chromosome numbers occur in the genus. The numbers known so far cannot be arranged in any polyploid series. Neither does the chromosome morphology suggest simple polyploidy conditions. The type of idiogram agrees very closely with the conditions in certain other Liliaceous genera, where long, medium and very small chromosomes are mixed in the same idiogram. The species dealt with in this paper represent only one of the 8 sections of the genus. Their idiograms must be regarded as rather derived. It is possible that the study of the other sections of the genus will reveal species with more primitive idiograms and in this manner furnish a clue as to the evolution of idiograms within the genus *Colchicum*. Such a study is planned, parallel with a study of the reactivity behaviour to colchicine within the whole genus of *Colchicum* and neighbouring genera.

Svalöf, January 13th, 1940.

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CYTOLOGICAL STUDIES OF DIPLOID AND TRIPLOID POPULUS TREMULA AND OF CROSSES BETWEEN THEM

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I. INTRODUCTION.

IT was stated by BLACKBURN and HARRISON (1924) that the diploid chromosome number is 38 and the haploid number 19 in *Populus tremula*; these numbers were confirmed by MÜNTZING (1936) and WETTSTEIN (1937). *P. tremula* has the chromosome number 2×19, in common with all other *Populus* species, about fifteen, the chromosome numbers of which are known (BLACKBURN and HARRISON, 1924; MEURMAN, 1925; WETTSTEIN, 1937; PETO, 1938).

The only reported exception has been recorded by BLACKBURN and HARRISON (1924), who consider *P. balsamifera* to be a tetraploid; MEURMAN found (1925), however, the chromosome number $2x = 19$ for this species too.

MÜNTZING (1936) has shown that a clone of *Populus tremula* with gigas properties detected by NILSSON-EHLE (1936) was triploid with $2n = \pm 57$. Two further clones from other localities in Sweden, having morphological features similar to the previously known triploid clone, were shown by TOMETORP (1937) to be approximately triploid. The morphology of one of these clones is described by BLOMQUIST (1937). In Canada, PETO (1938) has reported the occurrence of triploid *P. canescens* and *P. alba*. NILSSON-EHLE (1938) has described the occurrence of a tetraploid seedling in the progeny of a cross $2x \times 3x$ *P. tremula*, and some· data concerning these crosses are given by BERGSTRÖM (1940).

In the present paper some results will be discussed, mainly concerning the meiotic behaviour of diploid and triploid *P. tremula* and the fertility and chromosome numbers of the progeny of crosses between diploids and triploids. The work is being carried out from a breeding point of view at the Institute for Breeding Forest Trees at Svalöf, Sweden.

II. MATERIAL AND METHODS.

The original material for the study of meiosis, as well as the crosses made, was collected from wild populations in Sweden. These parental clones are in the main selected by Dr. N. SYLVÉN, Svalöf, and spread all over the country. In the case of triploids their detection is due to reports of extremely large-leafed aspens made by several persons.

In the winter cut twigs were placed in greenhouses, fixations of pmc were made at the proper time, and crosses were performed according to the method described by WETTSTEIN (1937). Pmc:s were prefixed in Carnoy for a short time and then fixed in diluted chrome-acetic-formalin. The sections were cut to a thickness of 14 μ and stained in gentian violet. In some cases the somatic chromosome number, too, was determined for the parent clones, and then in root tips from suckers, planted in pots, or — in the case of some females — in the carpels.

On determining the 2n-number of seedlings the plants were planted in pots, and root tips fixed in diluted chrome-acetic-formalin. Sections, 10 μ thick, were cut and stained in gentian violet. In this connection some observations made as to the most suitable time for fixation may be mentioned briefly. Three series of fixations of the same plant material were made on different days under different weather conditions. The fixations were performed every other hour of the 24 hours. It appeared that fixations made at noon in sunlight gave numerous divisions, but the divisions were unclear without distinct metaphase plates. On the other hand, fixations made in the early morning or on cold, rainy days yielded few divisions but with pronounced metaphase plates, more suitable for counting. This result may be said to agree with the observations of KIHARA that an artificial cooling of the plant before fixation gives shorter chromosomes, more evenly distributed in the plate. On account of this superior result of fixation early in the morning the whole of the present material was fixed at that time.

In measuring the diameters of pollen grains a 40 objective and a 7x eyepiece were used, one unit $= 3,8$ μ. In measuring the stomata lengths the same system was partly used, partly a 40 objective and a 15x eyepiece, one unit $= 2,4$ μ. The drawings were made with a camera lucida, using a 100 fluorite objective and a compensating 30x eyepiece, giving a magnification of $\times$ 6000, which is reduced to $\times$ 4100. The microphotographs were taken with a Zeiss »Standard» camera.

III. TRIPLOID ASPEN.

1. OCCURRENCE.

At present 9 different clones of triploid aspen are known in Sweden. These clones are growing in the following different localities.

1. Lillö, Skåne, South Sweden (NILSSON-EHLE, 1936; MÜNTZING, 1936).
2. Våle, Medelpad, Middle Sweden (BLOMQUIST, 1937; TOMETORP, 1937).
3. Vittjärv, Norrbotten, North Sweden (TOMETORP, 1937).
4. Våle, Medelpad, Middle Sweden, in the neighbourhood of 2.
5. Vittjärv, Norrbotten, North Sweden, in the neighbourhood of 3.
6. Jansjö, Ångermanland, Middle Sweden.
7. Häggnäs, Västerbotten, North Sweden.
8. Rosinedal, Västerbotten, North Sweden.
9. Vänsjärv, Norrbotten, North Sweden.

In most cases the 2n- as well as n-numbers have been determined, and the triploid behaviour is recorded by crossing with diploid partners. In no cases, however, have the chromosome numbers been ascertained to be exactly 57, but may be assumed to be ± 57; a difference of one chromosome may be possible but rather unlikely. In Fig. 18 a somatic plate with 57 chromosomes is drawn.

2. CELL SIZE.

The criteria adopted when searching for triploid aspens were extremely large leaves. MÜNTZING (1936), TOMETORP (1937) and BERGSTRÖM (1940) state that triploid aspens have longer stomata than

TABLE 1. *Size of the leaves, lengths of stomata and chromosome number in wild P. tremula.*

Leaves	2n	Lengths of the stomata										Mean	No. of trees
		6,6—7,0	7,1—7,5	7,6—8,0	8,1—8,5	8,6—9,0	9,1—9,5	9,6—10,0	10,1—10,5	10,6—11,0	11,1—11,5		
Small	38		2	4	1							7,7±0,13	7
»	—	5	38	63	19							7,7±0,03	125
Large	38				1	1					1	9,5±0,93	3
»	57				4	9	1	1				8,7±0,10	15
»	—			4	3	7	1					8,5±0,13	15

diploid ones. In Table 1 the correlation between size of the leaves, length of stomata and chromosome number are shown for the present

material, which also includes the triploids of MÜNTZING and TOMETORP. In estimating the length of stomata, 50 stomata from each of three leaves from every tree were measured. One of the three leaves was extremely large, one medium and one small, but all belonged to the larger leaves of the tree. The length of the stomata vary much within the leaf; consequently the calculated mean, tabulated as characteristic of the tree, cannot be of greater accuracy. 1 unit is equal to $3{,}8\,\mu$. It will be seen that all small-leafed trees have short stomata and as far as known $2n = 38$. The large-leafed trees have on an average larger stomata than the small-leafed ones, but as regards the chromosome number the large-leafed trees with long stomata are diploids as well as triploids. Taking the length of stomata as a measure of cell size one must conclude that *the size of the leaves depends on the size of the cells. The size of the cells in turn depends on chromosome number* (triploids having larger cells than most diploids) *and on other presumably hereditary factors not known* (there are diploids having as large cells as the triploids and even larger). That there is a correlation between size of the leaves and stomata length (cell size) in the individual tree, too, is illustrated by the means of the stomata length of the respective large, medium and small leaves of all the 165 trees measured.

Size of the leaves:	large	medium	small
Mean length of the stomata:	7,90	7,88	7,79 units

Thus the easily recognizable variation in leaf-size within the tree is associated with a variation in cell size, and in consequence *the cell size is influenced by modifying factors, too. Thus the length of the stomata as a measure of cell size in aspen is of no higher value as an indicator of triploidy than the leaf-size per se.* However, the leaf-size is of value in searching for triploid aspens as a means of eliminating non-triploid trees. For — in addition to the small number of small-leafed trees, which have been 2n-determined — a great number of such trees has been used in crossing, and all of them have given perfectly uniform progenies; not a single one has given the variable offspring, characteristic of triploid parents. Thus it seems rather unlikely that small-leafed triploid aspens exist.

3. POLLEN PROPERTIES.

MÜNTZING (1936) and TOMETORP (1937) have shown that triploid aspens have larger pollen grains and poorer pollen than diploid ones. Working with triploid *P. alba* and *P. canescens* and with diploid species and hybrids with meiotic disturbances, PETO (1938), however, arrived

at the conclusion that in his material no relation exists between either chromosome numbers or meiotic behaviour and size of the pollen grains and quality of the pollen.

Data relating to the pollen size and quality of the present material are given in Tables 2 and 3. Table 2 shows the diameter of the pollen grains. Every measurement comprises 100 pollen grains, 1 unit $= 3{,}8\ \mu$

TABLE 2. *Size of the leaves, chromosome number and diameter of the pollen grains in wild P. tremula.*

Leaves	2n	Diameters of the pollen grains						Mean	No. of trees
		9,1—9,5	9,6—10,0	10,1—10,5	10,6—11,0	11,1—11,5	11,6—12,0		
Small ...	38		4	3	6			10,5 ± 0,11	13
» ...	—	3	18	14	4	1		9,9 ± 0,07	40
Large ...	38				1			10,8	1
» ...	57			2	5		1	10,8 ± 0,13	8
» ...	—	1	3	2	2			9,8 ± 0,18	8

The small-leafed trees with unknown 2n-numbers may for very good reasons be considered as diploids. Then it is obvious that the triploid aspen has, on an average, larger pollen grains than the diploid one.

TABLE 3. *Size of the leaves, chromosome number and per cent good pollen in wild P. tremula.*

Leaves	2n	Percentage of good pollen														Mean
		30	35	40	45	50	55	60	65	70	75	80	85	90	95	
Small ...	38			1		1	1	1	3	5	1			2	5	74,0 ± 4,2
» ...	—						1		2	3	6	5	5	6	8	76,5 ± 1,7
Large ...	38											1			1	87,5
» ...	57	1			1	1	1			2	2					58,8 ± 4,7
» ...	—						1				1	3	1			75,8 ± 4,4

The two populations, however, overlap each other. Consequently there is no possibility of ascertaining whether the large-leafed trees with unknown 2n-numbers are diploid or triploid with the aid of the pollen diameter. MÜNTZING (1936) presents a bimodal curve for the pollen diameter of triploid aspens. In the present material no trace of such bimodality has been observed, but occasionally bimodality may occur (see below and Fig. 13). Table 3 gives the quality of the pollen. Every estimate is as a rule based on about 200 pollen grains. However, there

is no sharp boundary between »bad» and »good» pollen. — MÜNTZING works, obviously for that reason, with three categories: »good», »dubious» and »bad» grains. — Owing to this fact the estimates must be considered to be rather uncertain. Furthermore, the sampling error of each estimate must be high. There is a wide range of variation in percentage of good pollen among the trees of every category. Of course this variation is partly due to errors, but undoubtedly there is a significant variation, too. This variation may be due to external or internal factors. On an average, however, the triploids have poorer pollen than the diploids. But as in the case of the pollen grain diameter the variations overlap.

The inference must be that *the triploids have large pollen grains and poor pollen*, but many diploids have as large pollen grains and as poor pollen as many triploids — at least when the estimates are based on only one or a few samples.

However, the distinction between diploid and triploid pollen is somewhat more sharply defined, when both diameter and quality are considered together. Further, in the triploid pollen there is sometimes one or other elongated, sometimes constricted grain. Thus the pollen sample has a limited value in determining whether an aspen is diploid or triploid.

4. SEX.

Of the reported nine triploid aspens four are pure males, three pure females and two the sex of which is unknown. Nothing is known about the sex determining mechanism of the diploid aspen. But assuming this to be the type of a monofactorial back-cross, we may for convenience denote the one sex with XY and the opposite sex with XX. The triploid aspen must have originated as a result of a fusion between one unreduced and one reduced gamete, as MÜNTZING (1936) points out. Then if the unreduced gamete is of the XY-sex, the result must be one XXY-triploid. If, on the other hand, the unreduced gamete is of the XX-sex triploids of two constitutions may be produced, viz. XXX and XXY. Thus we have two different constitutions of the triploids as far as sex determinators are concerned. If the frequency of unreduced gametes is the same in both sexes of the diploid, and the chance of surviving is the same for both male and female unreduced gametes, the numerical relation between the two triploid types should be 3 XXY : 1 XXX.[/] The XXX-type must be expected to be of one pure sex. The XXY-type may be of the same or the opposite sex or an intersex. Now we have pure males and pure females among the triploids, but no

intersexes. Thus one of the triploid sexes may be assumed to have the constitution XXX and the other XXY. This is in agreement with WARMKE and BLAKESLEE's (1939) result on triploid *Melandrium*, i. e. 3A + XXX individuals are females and 3A + XXY individuals are males. However, one 3A + XXY-plant was actually an intersex.

IV. MEIOSIS OF DIPLOID ASPEN.

In their study of the meiosis of diploid aspen, BLACKBURN and HARRISON (1924) point out that the chromosomes were unequal in size: ›Nine small ones of more or less uniform size; nine others, larger than them, formed a graded series beginning with a member of just a little greater volume than the individual of the first group and ending with one more than four times its volume. Lastly there was a single chromosome . . . equalling in volume if not exceeding that of any two of the other eighteen›. This description is, indeed, very striking, especially the occurrence of one bivalent much bigger than the others (cf. MÜNTZING, 1936). In Fig. 1 a IM in polar view is drawn, showing this quite clearly. It is impossible to determine the bivalent condition of all bodies in polar view. In Fig. 9 a photograph of a IM is shown. Later on meiosis behaves quite schematically. BLACKBURN and HARRISON (1924) assume that there is a pair of heterochromosomes present in *P. tremula*. At any rate the difference cannot be great between the members of this pair, and as far as my experiences go, it will be exceedingly difficult to ascertain the presence of such a pair with but slightly differentiated members in *P. tremula*. The bivalents are small and numerous, in side view for the most part intensely crowded. The occurrence of occasional bivalents, asymmetrical in shape, may be due to chance.

For some other *Populus* species heterochromosomes have been reported by MEURMAN (1925) and BLACKBURN (1929). PETO (1938). treating *P. tremuloides,* heterochromosomes of which are reported (ERLANSON and HERMANN, according to PETO), states, however, that ›no definitely heteromorphic pair has been found consistently›.

Out of 16 clones, the meiosis of which has been studied, 12 behaved quite normally, but the remaining four showed a varying number of chromosome bodies in polar view of IM, ranging from 19 to as many as 31. Thus in Fig. 2 there are 20 quite clear ›separate bodies›, while in Fig. 3 there are 26. No doubt, these clones, too, have the exact diploid chromosome number. For at all events some IA and IIM plates
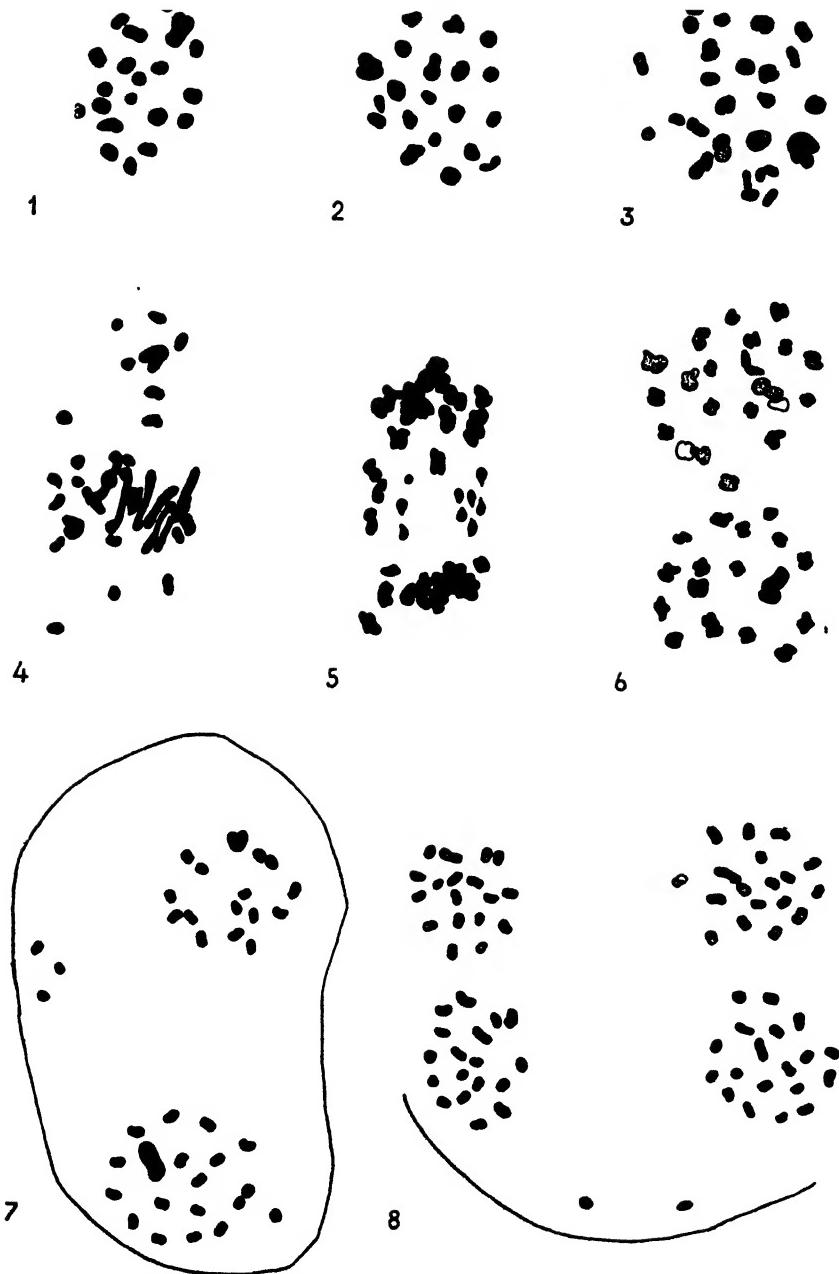
Figs. 1—8. Meiosis with univalents in diploid *P. tremula*. — Fig. 1, IM with 19 bivalents; Fig. 2, IM with 20 separate bodies, bivalents and univalents; Fig. 3, IM with 26 separate bodies, bivalents and univalents; Fig. 4, IM in side view, with 8 bivalents and 22 univalents; Fig. 5, IA with lagging univalents in division; Fig. 6, IA in polar view (the two plates separately drawn) with 18—19—2 chromosomes; Fig. 7, IIM with 19—16—3 chromosomes; Fig. 8, IIA with 19—18 : 19—18 : 2 chromosomes (the sister plates separately drawn).

have shown the sum of 38 chromosomes.   In side view of IM, Fig. 4 and photograph, Fig. 10, it is quite clear that only bivalents and univalents occur (two of the univalents are evidently the members of the large chromosome pair).   Thus Fig. 2 should represent $18_{II} + 2_I$ and

TABLE 4.   *The frequency of univalents in some diploid aspens.*

Origin of the clones	Number of separate bodies per pmc													Mean	% good pollen
	19	20	21	22	23	24	25	26	27	28	29	30	31		
Östergötland	1	5	5	2	0	1								20,9	70,0
Uppland	6	2	4	1	0	0	1							20,4	38,9
Medelpad.....	4	2	1	0	1									20,0	80,3
Hälsingland	1	2	3	3	1	0	2	2	1	0	0	0	1	23,2	67,1

Fig. 3 $12_{II} + 14_I$.   In Table 4 some figures are given, showing the degree of asyndesis of the four clones.   It will be seen that the frequency of univalents is especially high in the clone from Hälsingland.

At IM the univalents are distributed irregularly in the cell (Fig. 4

Figs 9—11   Meiosis in diploid *P tremula* — Fig 9, IM in polar view with 19 bivalents; Fig 10, IM with univalents in side view, Fig 11, IA in side view with lagging univalents

and photograph, Fig. 10).   Frequently some are situated in the polar regions.   At IA a larger or smaller number of bodies, certainly univalents, lag behind and divide or begin to divide (Fig. 5 and photograph, Fig. 11).   Fig. 6 shows a IA in polar view (the plates are drawn separately).   In one plate there are 18 chromosomes, in the other 19 and another 2 are situated at one of the poles.   This IA must have been

brought about by division of one univalent and may be interpreted as the result of a IM with $17_{II} + 4_I$, one univalent of which has divided, and the halves passed to opposite nuclei, one univalent has been included undivided in the one nucleus, and the other two univalents have remained undivided in the polar region outside the plate. These two univalents may or may not be included in a nucleus at interphase. In most cases the lagging bodies do not arrive at the poles before the first division is finished, and the cells enter the resting stage. Evidence of such an elimination is shown in Fig. 7 (a IIM) where two plates with 19 and 16 chromosomes respectively are seen, and three more chromosomes are situated between the nuclei and close to the inside of the cell wall. Probably these chromosomes are undivided univalents, for the sum of the chromosomes is exactly 38. In Fig. 8 two chromosomes opposite to each other near the wall are situated apart from the plates. A univalent may have passed through the first division undivided, not being included in any of the nuclei and then passes through the second division separately. In this figure (the sister plates are drawn separately) one of the two pairs of plates at IIA has 19 and the other 18 chromosomes. This fact may be understood if each of the daughter plates from first division has received 18 split chromosomes (from bivalents or univalents) and one univalent which has split later on.

Thus, in *Populus tremula* univalents divide or do not divide at first division. The split or unsplit univalents are included in interphase nuclei or are left outside them. At second division split chromosomes, included in a nucleus, pass to one of the two nuclei without further division. The split chromosomes, excluded from the daughter nuclei at IA can finish the second division separately. Indications of asynaptic behaviour in diploid *P. tremula* were reported by MÜNTZING (1936). In Table 4 estimations of pollen quality are given for the asynaptic aspens. The mean percentage of good pollen of the other 12 clones with quite regular meiosis was 79,9. Thus it is seen that the variation in pollen quality in diploid aspen can to some degree be attributed to meiotic disturbances.

With regard to the occurrence of univalents in diploid aspen several causes may be taken in consideration. Structural hybridity seems very unlikely. In Sweden no other wild *Populus* species than *P. tremula* occurs. That structural changes within the species should take place, involving so many chromosome pairs and without multiple associations, seems hardly plausible. Genes causing an omission of chiasma formation are known in several plants (maize, BEADLE and MC CLINTOCK,

1928; *Datura*, BERGNER et alii, 1934; *Alopecurus*, the present writer, in press). The most probable cause, however, is to be sought in external influences. STRAUB (1937) has shown that changes in temperature at leptotene and pachytene in *Gasteria trigona* affect the chiasma formation and result in a varying number of univalents at IM. Pmc:s of examined aspens were fixed from twigs, which only a few days previously (in January and February) had been cut out-of-doors and transferred to a greenhouse. Probably pmc:s of some clones were at that very moment in a sensitive stage. Outdoor pmc:s pass through meiosis in early spring at low temperature. No doubt considerable changes in temperature may sometimes occur in nature at that time. Thus if the occurrence of univalents in aspen is connected with changes in temperature univalents will also arise in nature. On the other hand, an occurrence of univalents in high frequency will furnish special opportunities for the formation of unreduced gametes. Owing to the fact that triploid aspen is not especially rare, unreduced gametes must be regarded as fairly common in nature.

V. MEIOSIS OF TRIPLOID ASPEN.

A description of the course of meiosis in the triploid male aspen from Skåne has been given by MÜNTZING (1936). This author has established that IM of the triploids is characterized by a varying number of trivalents and univalents in addition to bivalents, and that chromosomes are often lagging at IA and may then constitute a connection between the nuclei, which may give rise to unreduced gametes. Further, he has proved that irregularities may occur even at the second division. — The present writer has studied meiosis of the same triploid as MÜNTZING, and in addition the meiosis of two triploid male clones growing in Medelpad in the neighbourhood of each other and also the meiosis of a few available emc:s of the triploid female clone from Norrbotten. In all cases examined meiosis was of the same appearance as that described by MÜNTZING. On account of the small and rather numerous chromosomes it is impossible to give any detailed statistics with respect to the frequency of trivalents, the amount of chromosome elimination, chromosome numbers of the first and second anaphase and so on. On the whole, the formation of interphase nuclei and tetrads seems to proceed rather regularly and the resulting young pollen grains appear uniform in size, having one nucleus (photograph, Fig. 12). However, most of the pollen grains contain chromatin bodies outside

the nucleus. These bodies are probably derived from lagging and eliminated chromosomes, which almost invariably can be seen in the first as well as in the second anaphase. The pollen nuclei must be presumed to have varying chromosome numbers. Infrequently a loculus in one or other stamen of a flower presents a quite different picture, as illustrated by the photograph, Fig. 13. In these cases the pollen grains are of very different size, some being normal-sized, some much larger. The large pollen grains may have one, two, three or four nuclei. These facts make it clear that meiosis and the formation of cell walls are influenced by external or internal factors, affecting
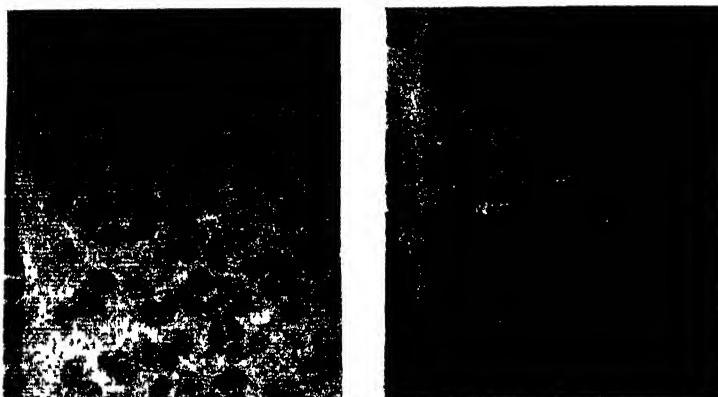
Figs 12—13. Young pollen grains of triploid *P tremula* — Fig 12, one loculus with uniform grains of normal size; Fig 13, one loculus with unequal-sized grains, mainly large grains with one, two or three nuclei.

only parts of a flower. No doubt, these large pollen grains with one or more nuclei have higher chromosome numbers than is usually the case, and if they are capable of functioning they will give rise to offspring with more than the triploid chromosome number.

## VI. CROSSES BETWEEN DIPLOID AND TRIPLOID ASPEN.

In the winter of 1938 a number of crosses between diploid and triploid aspen were made according to the method described by WETTSTEIN (1937). As triploid parents the male triploid from Skåne and Medelpad, used by BERGSTRÖM (1940), and the female triploid from Norrbotten were employed. As diploid parents clones from different localities in Sweden were selected. A total of 29 crosses yielded a varying number of offspring plants.

The seedlings of these crosses show a very characteristic behaviour,

TABLE 5. *Chromosome numbers of the progeny of crosses between diploid and triploid P. tremula.*

Triploid parent	Cross number	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	±4x	Mean 2x–3x	%2x	n
♂ Skåne ...	62	23	5	2	3	4	1	2	5	6	2	7	7	4	8	3	7	4	4	4	1	1	45,9±0,67	23,5	99
» ...	200	2	0	0	0	4	0	0	1	0	0	1	0	2	0	0	0	0	0	1	0	13	46,4±2,51	28,6	20
» ...	203	11	5	1	3	0	1	0	1	0	0	2	1	1	0	0	0	0	0	1	0	4	41,0±0,81	42,3	30
» ...	215	9	0	1	3	0	1	2	0	1	3	2	1	4	0	0	1	1	3	2	0	0	45,7±1,08	26,5	34
» ...	254	15	4	2	4	8	9	8	4	9	17	19	10	16	26	8	8	4	3	1	1	0	47,2±0,35	8,5	176
» ...	293	6	1	0	1	2	2	2	1	2	1	8	3	7	5	5	9	1	4	0	2	0	48,5±0,20	9,7	62
» ...	S 10 numbers	13	2	6	2	3	0	0	2	5	3	7	6	3	2	0	1	2	2	2	1	0	45,0±0,73	21,0	62
Total ..		79	17	12	16	17	14	14	14	23	26	46	28	36	41	16	26	12	16	7	5	18	46,3±0,28	17,0	483
♂ Medelpad	297	7	0	3	1	0	3	1	1	0	1	2	2	1	0	7	1	1	0	0	3	0	46,5±1,12	20,6	34
»	304	1	0	1	0	0	0	0	0	0	0	0	0	1	0	2	0	0	1	0	0	0	47,8±2,88	16,7	6
»	311	3	1	0	3	0	1	0	2	0	1	1	2	1	0	2	0	1	0	1	0	0	45,8±1,33	15,8	19
Total ...		11	1	4	4	0	4	1	3	0	2	2	4	3	1	11	1	2	1	1	3	0	46,3±0,70	18,6	59
♀ Norrb....	184	2	0	3	0	0	1	0	2	3	3	2	2	2	0	2	0	0	0	0	0	0	45,8±0,92	9,1	22
»	223	2	1	1	0	0	1	1	0	1	4	1	2	3	1	1	2	2	0	0	0	0	46,7±0,98	7,7	26
»	230	2	0	0	0	0	0	0	2	0	0	0	0	0	2	0	0	1	1	0	0	0	47,1±1,97	22,3	9
»	S 7 numbers	3	1	1	0	1	0	1	0	1	1	5	0	2	0	0	0	1	1	0	1	0	45,8±1,27	16,7	18
Total ...		9	2	5	0	4	2	2	4	5	9	8	4	7	3	3	2	4	2	0	0	0	46,2±0,57	12,0	75
Total		99	20	21	20	21	20	17	21	28	37	56	36	46	45	30	29	18	19	8	8	18	46,3±0,23	16,5	617
Calculated		0	0	0	1	4	13	31	58	86	106	106	86	58	31	13	4	1	0	0	0	0	47,5	0,0	599
Total, surviving plants, Aug. 1939...		88	18	15	15	19	16	14	15	20	27	47	25	39	43	23	25	15	15	7	7	15	46,3±0,20	17,8	508
% loss		11,1	9,0	28,6	25,0	9,5	20,0	17,7	28,6	28,6	27,0	16,1	30,6	15,2	4,4	23,3	13,8	16,7	21,1	12,5	12,5	11,8±2,2	19,0±0,1	0,3	

0,3

compared with seedlings from crosses between diploid parents; the
latter are uniform in size and appearance (Fig. 14), the former varying
in size, in the shape and thickness of their leaves and so on (Fig. 15).
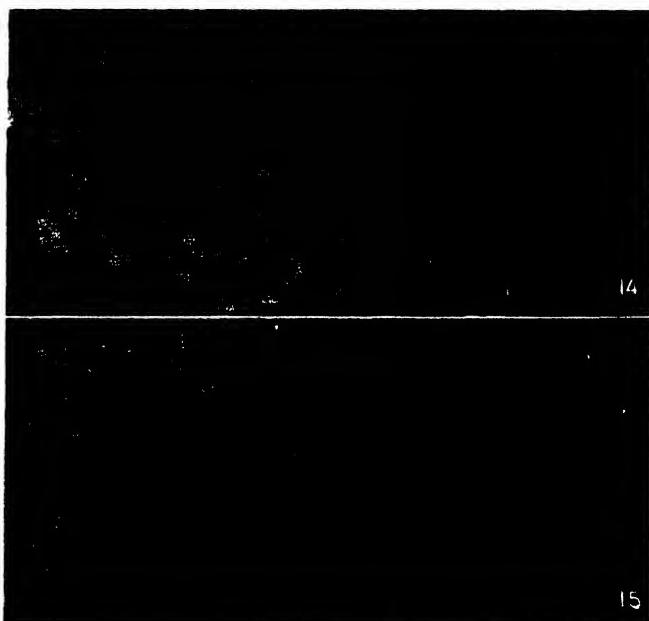
Figs. 14—15. Seedlings of *P. tremula*. — Fig. 14, Progeny of a cross 2x — 2x aspen;
Fig. 15, Progeny of a cross 2x — 3x aspen.
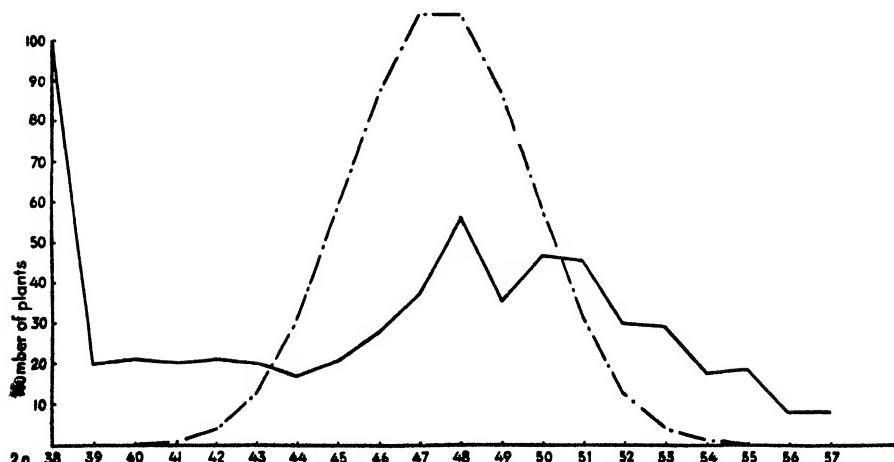
Fig. 16. Diagram showing the experimental distribution of the 2n-numbers in
crosses between diploid and triploid *P. tremula* (———) compared with the binomial
distribution (—·—·—).

The chromosome numbers have been determined without selection for as many plants as possible in the offspring, and the result is shown in Table 5. These chromosome numbers, however, are not determined with absolute accuracy; the error is on an average ± 1 chromosome. The reason for this is the great difficulty of making an accurate count of the somatic chromosome numbers (cf. MÜNTZING, 1936 and BERGSTRÖM, 1940) owing to the fact that practically in all plates the very small chromosomes, at least at some point, are crowded, making a clear distinction impossible, or the median constriction of some chromosome may be long and uncoloured, so that this chromosome might have been counted as two. In the determination of a chromosome number
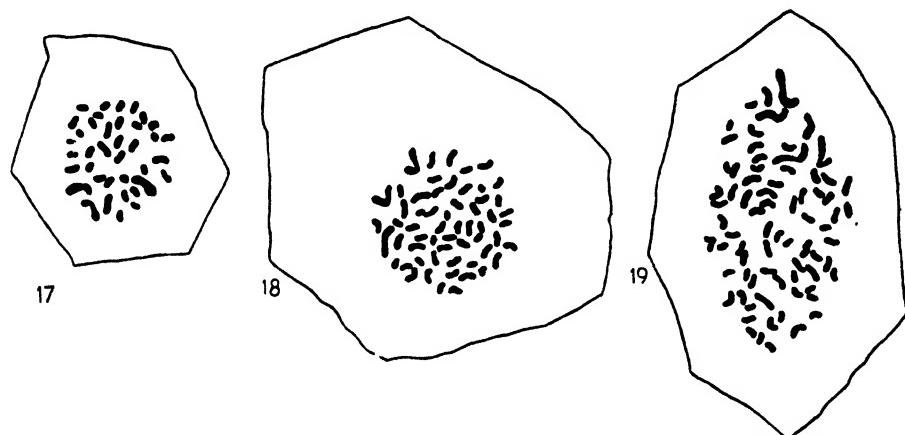
Figs. 17—19. Somatic plates of *P. tremula*. — Fig. 17, 2n = 38; Fig. 18, 2n = 57; Fig. 19, 2n = 76.

the three best plates were counted, and when the results have differed by one, two or occasionally more chromosomes the mean was taken. In Figs. 17—19 some somatic plates are drawn.

One of the most striking features is that all chromosome numbers between 38 and 57 are represented (cf. BERGSTRÖM, 1940). In general, in crosses between diploids and triploids the intermediate chromosome numbers are very rare, if represented at all, in the progeny (DARLINGTON, 1937; UPCOTT and PHILP, 1939). On the other hand, almost all offspring plants have the diploid or a slightly higher chromosome number. That is true, even when the data of a large progeny are given (BELLING and BLAKESLEE, 1922, on *Datura*). But recently UPCOTT and PHILP (1939) were able to show that the progeny from 3x — 2x crosses in *Tulipa* had the intermediate 2n-numbers very well represented, and

the authors state: »These data provide the only example of triploid-diploid progeny with chromosome numbers evenly distributed between those of their parents». However, similar results have been previously reported, viz. between diploid and triploid apple. The data were recently summarized by WANSCHER (1939). This case is according to DARLINGTON (1937) a very special one and is considered by him to accord with the view he arrived at in other ways, that the x-number 17 in *Pyrus* is a secondary one.

The mean 2n-number for the whole material of 599 plants, ranging between 38 and 57, is $46,3 \pm 0,23$. Different crosses have, of course, a somewhat different mean, but in view of the standard errors the differences are not significant, perhaps with the single exception of cross No. 203, which seems to have a significantly lower mean value than the others.

MATHER. (1935) states that the odd chromosomes in triploid *Triticum* at meiosis are distributed at random and in consequence gametes must be produced with binomial frequencies with n-numbers ranging between x and 2x. UPCOTT and PHILP (1939) have worked with the chromosome numbers of the pollen grains in *Tulipa* and summarized the literature, and they show that this law seems to have a general application. The mean n-number of the pollen-grains is $\dfrac{3x}{2}$ or slightly below, according to the elimination of chromosomes at meiosis. There are no reasons for believing that 3x-aspen behaves in a different manner. Consequently the progeny of crosses diploid—triploid aspen should have $\dfrac{2x + 3x}{2}$, i. e. 47,5 chromosomes on an average; in fact, the values is $46,3 \pm 0,23$ or somewhat lower than expected. This slight difference may be inferred as being due to chromosome elimination at meiosis or to the limited accuracy with which the chromosome numbers are determined, or to both facts in co-operation. However, another effect of a random distribution of the univalents at meiosis is that the 2n-numbers of the progeny must also be distributed in binomial frequencies. The binomial frequencies of 599 variants are given in Table 5. No doubt, the calculated and the experimental values do not agree at all. This fact is further illustrated in the diagram, Fig. 16. In spite of the fact that all intermediate 2n-numbers are realized and that the mean 2n-number is only slightly below the calculated, the distribution differs very much. The intermediate · 2n-numbers, from 44 to 50, are deficient and the extremes,

38—43 and 51—57, are in excess. Especially the excess of 2n = 38 is very great, 99 plants or 16 % having this 2n-number. According to the law of chance only 1 plant in 524,288 should have had this 2n-number, i. e. very likely none of the present 599 plants. The percentage of 2n = 38 plants varies between different crosses within rather wide limits, being only 7,7 % for Cross 223 and 42,3 % for No. 203.

The general explanation of the discrepances between calculated and experimental values in crosses of this type is that elimination of gametes or zygotes or both takes place (cf. DARLINGTON, 1937). Probably this is true of the aspen crosses too. There are even some data indicating the course of this elimination. At meiosis a certain amount of chromosome elimination occurs; this may imply a tendency to decrease the 2n-numbers of the offspring. As shown in Table 3, the triploids have on an average only 58,8 ± 4,7 % morphologically good pollen grains, varying between 30 and 75 % for different clones and even varying as much as 50 % within the same tree; this variation may be due partly to sampling errors, partly to real differences. Even within the diploid aspen the percentage of good pollen grains varies, but for the best trees it is almost 100 %, and on an average higher than for the triploid.

This must indicate a certain amount of gamete elimination occurring in the triploid male aspen. This elimination may be calculated at about 40 % of the pollen grains.

TABLE 6. *Number of seeds per capsule and germination percentage in crosses 2x × 2x and 2x × 3.x.*

2x — ♀ No.	Seeds per capsule		Germination %		Number of crosses	
	× 2x	× 3x	× 2x	× 3x	× 2x	× 3x
3	12,0	6,5	100	62	3	1
26	10,1	5,3	94	90	3	1
47	7,3	3,7	98	98	2	1
54	8,5	5,4	95	98	6	3
100	9,5	6,1	100	90	3	1
101	12,2	3,7	78	91	1	1
Mean and total...	9,9	5,1	94	88	18	8

In Table 6 the seed-setting per capsule and the germination percentage of the seeds are given for crosses between the same mother trees and diploid and triploid males respectively. In estimating the

number of seeds per capsule the seeds of 10 mature capsules were counted under the stereomicroscope with a magnification of 10 times, and the mean was calculated. The tabulated values are then the mean of all crosses with this mother tree; the number of crosses are given on the left side of the table. All six mother trees produced considerably fewer seeds in crosses with triploid males than with diploid ones. The average values are 9,9 seeds for 2x—2x crosses and 5,1 seeds for 2x—3x crosses, that is, an elimination of 52 %. Moreover, the seeds of the 2x—3x crosses did not in general germinate as well as the seeds of the 2x—2x crosses, the mean being 88 and 94 % respectively. The difference is, however, not significant. The main cause of this lack of significance is probably the great sampling error, for the germination tests were made with 1×100 seeds only. — At any rate it may be stated that there is an elimination of at least 50 %. This elimination is probably entirely or to the greatest extent an elimination of zygotes and embryos. That some ovules were not fertilized owing to insufficient pollen tube growth seems to be unlikely; the pollination was performed with a brush and every stigma was powdered with a large amount of pollen grains. — This fertility, reduced to about 50 %, must at any rate be regarded as surprisingly high. UPCOTT and PHILP (1939; cf. DARLINGTON, 1937) state that it is possible »to predict the fertility of a given 2x—3x cross from the 2n-numbers of the parents». They argue that the majority of 2x—3x offsprings are 2x-plants, and that the proportion of x-pollen grains depends upon x. In the case of aspen x is $= 19$, and only 1 pollen grain in 524,288 is haploid. Consequently, the fertility ought to be very low, but on the other hand the fertility is rather high, the reason being that the aspen is an exception to the law »that the majority of 2x—3x offspring is 2x-plants». The variation in percentage of 2x-plants in different crosses may consequently be inferred as being due to different viability of descendants with intermediate 2n-numbers. Then a high percentage of 2x-plants should be connected with low fertility. Indeed, this percentage is high just in those crosses where the number of counted plants is small. The number of 2n-determined plants, in turn, will be rather strongly correlated to the largeness of the progeny, i. e. to the fertility of the cross.

In Table 7 seed-setting and quality for reciprocal crosses 3x—2x are given. The average seed-setting is only 3,9 seeds per capsule, compared with 5,1 for the 2x—3x crosses; further, the germination percentage is reduced to 58. It is not likely that the 3x females have fewer ovules than the 2x ones, rather the contrary, for the triploid capsules

are larger than the diploid ones. Then the reduced fertility has to be attributed to elimination. This elimination must be the sum of gamete- and zygote-embryo-elimination. Assuming that the gamete- and zygote-elimination is of the same magnitude in 3x—2x crosses as in 2x—3x crosses it is possible to calculate an expected fertility for the 3x—2x crosses, based on the values of the 2x—3x crosses. Starting with 9,9 as the potential number of ovules, there is at first 40 % gamete elimination. 5,94 ovules remain to be fertilized, 47,5 % of these are eliminated as zygotes or young embryos. The germination percentage was 88 %, i. e. a further elimination of 12 % takes place as an elimination of old embryos, and thus the final value will be 2,75 young seedlings per capsule. The experimental value was 3,9 seeds per capsule, 58 % of which germinated, i. e. 2,26 seedlings per capsule. Thus

TABLE 7. *Number of seeds per capsule and germination percentage in crosses 3x $\times$ 2x.*

3x — ♀ No.	Seeds per capsule $\times$ 2x	Germination % $\times$ 2x	Number of crosses $\times$ 2x
67	3,3	65	1
56	4,4	50	7
Mean and total	3,9	58	8

the experiment gave a somewhat lower value than that calculated, but this may easily be due to the rather high error occurring in the estimation of good pollen (58,8 $\pm$ 4,7 %) and, of course, all the other values, too, have more or less sampling errors. Thus it seems that *the gametic abortion is of about the same magnitude in both sexes of the triploid aspen and that the elimination of zygotes and embryos is of about the same magnitude in both the reciprocal crosses between diploid and triploid aspen.* This result agrees with the *Pyrus* crosses (NEBEL, 1933; cf. WANSCHER, 1939), but is contrary to the bulk of diploid, triploid crosses (cf. DARLINGTON, 1937), where the 3x—2x direction yields a somewhat wider range of variation in 2n-number of the progeny than the 2x—3x direction does.

However, there is a striking difference between the germination percentage of reciprocal crosses. When the triploid is the male parent, the seeds germinate to 88 %, but when the triploid is the female part the germination percentage is only 58 %. This may indicate that the

elimination of embryos takes place later in 3x—2x crosses, the ovules having had time enough to develop so far as to be counted as seeds before the embryos die.

A further plant elimination occurred before the fixations were made. The seeds were sown in boxes, after about four weeks all seed-lings or some of them were transplanted into other boxes — without selection — in order to give more space to the plants. After another four-week-period the plants were transplanted again, now to pots in order to produce root tips for fixation, and as many of these plants as yielded roots were fixed. In all these instances plant elimination occurred. In the boxes a large amount of the seedlings died, mainly because of attacks by fungi, and later some plants died with or without

TABLE 8. *Plant elimination in offspring of crosses 2x × 3x.*

Plant categories	Number	%
Planted in pots, spring 1938	2369	100
Fixed, summer 1938	962	40,6
Counted	617	26,0
Counted surviving, autumn 1939	508	21,4
Non-fixed + fixed but non-counted, surviving, au-tumn 1939	669	28,2
Total surviving, autumn 1939.........	1177	49,7

external symptoms. In Table 8 a survey is given of the plants planted in pots. Of the total of 2369 plants planted in pots only 962 or 40,6 % could be fixed, a small part of the remaining 59,4 % died before they had grown up sufficiently to be fixed; the greater part was alive, but grew so slowly that they did not produce any roots for fixations during the whole summer. Of the fixed 962 plants only 617 could be 2n-determined, the fixation of the rest did not give plates clear enough for counting.

Several of these different types of elimination may be considered to be numerically selective, especially the gamete abortion and the zygotic and embryonic lethality. Among the seedlings and young plants a certain degree of the elimination is probably numerically selective, another part of this elimination can be attributed to injuries due to transplanting and so on. In Tables 8 and 5 it will be seen that only 508 of the 617 plants counted survived the first winter, i. e. a loss of 17,7 %. The loss is 11,1 ± 0,3 % for the 2x-plants and 19,0 ± 0,1 %

for the intermediate plants (Table 5) and thus the percentage of 2x-plants has increased from 16,5 to 17,8. *It therefore seems to be true that in agreement with the results of other such crosses, the non-binomial distribution of the 2n-numbers in progenies of diploid and triploid crosses is due to a numerically selective elimination of gametes, zygotes and embryos, seedlings and young plants.*

The above cited *Pyrus* crosses, with a large amount of intermediate 2n-numbers in their offspring, are considered by MOFFETT (1931; cf. DARLINGTON, 1937) to show a higher viability of $17 + 7$ gametes than of others. However, as pointed out by DERMEN (1936; cf. WANSCHER, 1939), this view is wrong. Instead, the distribution agrees very nearly with the binomial distribution, which is to be expected if no numerical selection takes place. For example, only one occasional plant out of 241 had the 2x-number.

It is possible to divide the known cases of crosses between diploids and triploids into three groups according to the 2n-numbers of the offspring.

Group 1. The intermediate numbers are almost completely lacking, at least in the direction 2x—3x. The 3x—2x direction yields some intermediates but in very reduced frequencies. The highest 2n-number for this group is 12 (*Tulipa, Datura, Solanum*). To this group belongs the main part of all crosses between diploids and triploids.

Group 2. The intermediate numbers are well represented but in reduced frequencies. Reciprocal crosses give the same result. Only one case is known, *Populus tremula*, $x = 19$.

Group 3. The 2n-numbers of the offspring are present in binomial distribution between 2x and 3x. Reciprocal crosses give the same result. One known case only, *Pyrus malus*, $x = 17$.

The differences between these groups are, in fact, not qualitative but quantitative, consisting of a gradient viability of aneuploid plants. In trying to find a plausible explanation of the higher viability of aneuploids in Groups 2 and 3 a modification of DARLINGTON's view, concerning secondary polyploidy, may be attempted. Surely, the x-numbers 19 and 17 cannot be considered to be primary haploid numbers, but rather tetraploid ones, which are modified by duplication or loss of one or other chromosome, as DARLINGTON and MOFFETT (1930) have indicated for *Malus* and, of course, the primary genomes must be considered also to have been differentiated, structurally and genetically. Consequently the crosses belonging to Groups 2 and 3 are rather of a tetraploid, hexaploid type of crosses, contrary to the real (diploid, tri-

ploid) crosses of Group 1. Therefore aneuploid n- and 2n-numbers of *Populus* and *Malus* may imply smaller disturbances in the quantitative balance and more likely comprise all necessary types of chromosomes or chromosome structure than in the case of Group 1. Indeed, crosses between higher multiples in general show a higher fertility, greater frequency of intermediates, and better vitality of the aneuploids than crosses between lower multiples (MÜNTZING, 1937).

The difference between *Populus* and *Malus* may be more or less due to modifications. The progenies of *Malus* were raised under natural conditions by free or artificial pollinations. On account of the large seeds and seedlings it is easy to take good care of the offspring. The progenies of *Populus*, on the other hand, were raised from cut twigs. It is possible that this involves a greater demand on vigour for survival in both gametes and zygotes as well as embryos than under natural conditions, giving rise to the death of a greater number of subvital gametes, zygotes and embryos. Further, the seeds of *Populus* are small and very sensitive, the »struggle for life» will be relatively more severe in the germination and growth of the seedlings of *Populus* than those of *Malus*. However, there exists the possibility that real differences between *Populus*- and *Malus*-crosses occur.

In his report on meiosis of the triploid *Populus tremula*, MÜNTZING (1936) predicts the possibility of tetraploid individuals among the progeny of diploid, triploid crosses. The occurrence of a tetraploid plant in the progeny of a cross between diploid and triploid aspen has also been reported by NILSSON-EHLE (1938; cf. BERGSTRÖM, 1940). Approximately tetraploids even occur in this material, as seen in Table 5. Out of the total of 617 counted plants 18, or 2,9 %, had ± 4x chromosomes. The actual 2n-numbers have varied between ± 71 and ± 79 chromosomes, the exact tetraploid number being 76. For most of these plants the 2n-number is certainly exactly 76 (Fig. 19). But it is quite possible that some plants deviate slightly from the exact tetraploid number. That all these ± 4x-plants have originated as a result of the formation of unreduced gametes in the triploid parent is quite clear. The formation of unreduced gametes is also indicated by the studies of meiosis. That unreduced gametes need not necessarily have the exact somatic chromosome number has been pointed out by MÜNTZING (1937). All the 18 tetraploids were produced in crosses with the same triploid parent. But there are no reasons for assuming that the other two triploids used are not capable of producing tetraploids, too. These two triploids were used only to a small extent, as compared with the one pro-

ducing tetraploids, and only 3 out of 16 crosses with the latter produced tetraploids. The frequency of ± 4x-plants in the three crosses, having produced tetraploids, is very variable. In Cross 62 it is 1 tetraploid out of 99 plants, in Cross 203 it is 4 out of 30, and in Cross 200 it is 13 out of 20 plants. Such a variable frequency from 0 to a high value is also to be expected in view of the observation that an occasional loculus of the stamens produced large pollen grains in a very high frequency when other loculi produced pollen grains of normal size exclusively. Therefore the number of tetraploids may be correlated to the amount of the pollen grains used, derived from such exceptional loculi.

VII. STOMATA LENGTH AND PLANT HEIGHT OF THE PROGENY.

1. STOMATA LENGTH.

In Table 9 the stomata lengths are given for the aneuploid off-spring of the 2x—3x cross No. 257. The measurements were made at

TABLE 9. *Stomata-length and chromosome number of the aneuploid offspring of Cross 257, 2x—3x aspen.*

Chromosome number	38	39	40	41	42	43	44	45	46	47	48
Length of stomata ...	13,3	12,6	11,4	11,8	12,4	11,8	12,9	13,4	12,6	12,9	13,3
Chromosome number	49	50	51	52	53	54	55	56	Mean		
Length of stomata ...	13,8	13,2	13,9	11,9	13,0	11,7	12,5	14,5	12,8 ± 0,19		

the end of the second growing season. As a rule three plants of each 2n-number were measured and the mean calculated. In a few cases only two or even one plant was available for measurement. Of each plant 50 stomata of one leaf were measured — one unit being 2,3 μ. As seen, there is no connection between stomata length and 2n-number. The regression of stomata lengths on 2n-numbers, calculated according to FISHER's formula, is 0,05 with p > 0,9. The average stomata lengths of this aneuploid variation is 12,8 ± 0,19 units. Table 10 gives, for comparison, the mean stomata lengths of two pure diploid crosses, the aneuploid cross No. 257 of Table 8 and of the 2x—3x cross No. 200, distinguished by a large frequency of ± 4x-plants. No doubt, in spite of the non-

existent regression of stomata lengths on 2n-numbers within the aneuploid progeny, the mean stomata length of this variation, having the mean 2n-number 47, is significatively higher than the mean stomata length of the two diploid crosses. Furthermore, the mean stomata length of the $\pm$ 4x-plants is higher than the comparable values of the diploids as well as of the aneuploids. Thus the fact that the 2n-number is a factor influencing the stomata length (cell size), established by the treatment of diploids and triploids, also applies to aneuploids and tetraploids. And, as in the case of the diploids and

TABLE 10. *Stomata-length and chromosome number for diploid, aneuploid and tetraploid aspen.*

Cross number	301	276	257	200		
Chromosome number	38	38	38—56	38	50	$\pm$ 4x
Number of individuals	5	23	49	2	1	11
Stomata-length ..	11,6 $\pm$ 0,10	11,8 $\pm$ 0,15	12,8 $\pm$ 0,19	11,8 $\pm$ 1,25	13,0	15,3 $\pm$ 0,31

triploids, other factors also influence the stomata length of aneuploids and tetraploids. This is especially marked in the aneuploids, within which there is no regression of stomata length on 2n-numbers. Another factor operating here may be the chromosome components of a given 2n-number. A certain 2n-number may be built up predominantly by small or by large chromosomes, and that may influence the cell size. Further, genes may be assumed to exist, which influence the cell size, and these genes will be included in various ways with different combinations of the chromosomes.

2. PLANT HEIGHT.

The vigour of the 2x—3x offspring is very variable at the end of the second growing season. This variation is as great within as between the 2n-numbers. On an average the plants are poor (cf. BERGSTRÖM, 1940). In Table 11 measurements of plant height are tabulated for each 2n-number. The standard errors are great in agreement with the great variation in vigour among plants with the same 2n-number. The only conclusions to be drawn are that plants with 38 and 39 chromosomes are much taller than plants with any other 2n-numbers. The reason why the height of the 39-plants is so great is because some 38-plants may also have been included in this category owing to the

limited accuracy of the 2n-determinations. In general, plants having aneuploid 2n-numbers are inferior in vigour to diploid plants, as discussed by MÜNTZING (1937). One example quite comparable with the 2x—3x aspen offspring is given by MÜNTZING (1937) in the cross between the diploid (2n = 14) *Dactylis Aschersoniana* and the triploid hybrid between this species and its tetraploid derivative *D. glomerata*. In this cross one or a few plants of all intermediate 2n-numbers were realized. All the intermediate plants were much inferior in vigour as compared with the diploid *D. Aschersoniana*.

TABLE 11. *Chromosome numbers and plant heights after the second growing season in offspring of crosses* 2x × 3x *P. tremula.*

Chromo-some number	Mean height cm.	Max. height cm.	Number of plants	Chromo-some number	Mean height cm.	Max. height cm.	Number of plants
38	41,80 ± 3,16	139	84	50	22,21 ± 2,46	70	38
39	45,35 ± 5,77	86	17	51	25,88 ± 2,86	94	40
40	23,33 ± 2,31	44	15	52	24,67 ± 2,93	55	21
41	25,00 ± 4,02	58	15	53	33,65 ± 3,99	77	23
42	28,00 ± 4,41	80	16	54	24,54 ± 4,72	61	13
43	23,31 ± 4,35	66	14	55	25,15 ± 3,14	89	13
44	18,45 ± 3,02	40	11	56	32,83 ± 9,23	68	6
45	23,33 ± 3,41	50	15	57	33,20 ± 11,40	71	5
46	20,50 ± 2,20	61	16	•			
47	24,14 ± 2,47	63	22	•			
48	24,15 ± 2,94	97	39	•			
49	17,37 ± 2,68	54	24	± 4x ...	26,75 ± 3,74	57	16

A perhaps more surprising result is that the ± 4x-plants do not seem to be superior to the aneuploids in plant height. Here, however, another factor is involved. In Table 12 the mean plant heights of some progenies of pure diploid crosses are given. There are considerable differences in plant height of different crosses. The main source of these differences may be the occurrence of photoperiodic races within *Populus tremula*, which are very highly adapted to the special light climate of their habitats (SYLVÉN, unpubl.). All progenies have been cultivated at the institute at Svalöf, Skåne, South Sweden, at about 55° 42′ north latitude. Here the progenies of parents from North Sweden, as is the case of cross No. 204, develop very slowly. The same is to a high degree true of crosses between one southern and one northern parent, as exemplified by cross No. 301. Only crosses between

two southern aspens grow normally, crosses Nos. 251 and 288. With regard to the aneuploids it has been shown (Table 5) that the distribution of 2n-numbers is about the same for all crosses. Consequently, southern, northern and southern $\times$ northern crosses have contributed, on an average, equally to every 2n-number. Thus the means are comparable. But in the case of $\pm$ 4x-plants, 15 out of 16 measured plants occasionally belong to the Crosses 200 and 203, both of which have the triploid male aspen from Lillö at about 55° 42' north latitude as fathers, and as mother parents two different diploid females from Vittjärv in Norrbotten at about 66° north latitude, not far from the arctic circle.

TABLE 12. *Plant-heights after the second growing season of offspring of some 2x-crosses.*

Cross number	Combination	Mean height cm.	Max. height cm.	Number of plants
251	Belgium $\times$ Skåne	71,00 $\pm$ 4,84	125	20
276	Västergötland $\times$ Skåne	46,57 $\pm$ 3,20	91	42
301	Skåne $\times$ Medelpad	33,67 $\pm$ 2,79	99	45
201	Medelpad $\times$ Medelpad	37,47 $\pm$ 3,55	60	19
204	Norrbotten $\times$ Medelpad	20,84 $\pm$ 2,39	71	44
288	P. alba (Skåne) $\times$ Skåne	86,06 $\pm$ 8,90	168	18
286	P. alba (Skåne) $\times$ Norrbotten	42,47 $\pm$ 4,93	77	19

Consequently, the plant height of the $\pm$ 4x-plants is not comparable with the corresponding values for the other 2n-numbers. Further, nothing can be said as to the absolute vigour of tetraploid aspen, owing to the fact that the tetraploids, available at present, are not photoperiodically adapted to the actual light condition occurring at Svalöf.

VIII. SEX OF THE TETRAPLOIDS.

Of course, it has not yet been possible to determine the sex of the tetraploid seedlings produced. By means of grafting buds in the crowns of old fruitful aspens it will, however, be possible to determine the sex in the near future. In spite of this some conjectures as to the possible sex may be justified. According to the assumptions, made in the discussion of the sex of the triploids, the triploid male parents may have the constitution XXY. Then the diploid female must be XX. All the discussed tetraploids have originated as a result of unreduced

triploid male gametes, i. e. XXY-gametes. Therefore only one sort of tetraploids will result, viz. XXXY individuals. These may be males, females or intersexes. In *Melandrium*, WARMKE and BLAKESLEE (1939) found the 4A + XXXY individuals to be pure male in 65 cases and intersexes in 3 cases. If, on the other hand, the triploid male is of the constitution XXX, the diploid female must be XY. Then the tetraploids will be of two constitutions, viz. XXXX and XXXY. Both these constitutions may phenotypically be males, or the one male and the other female or intersex. Thus if the two tetraploid constitutions XXXX and XXXY are phenotypically different, the sexes of the tetraploids — in any case if tetraploids, derived from female unreduced gametes. are also available — will offer an opportunity to prove whether the male or the female represents the heterozygous sex in diploid *P. tremula*. If, on the contrary, both the constitutions have the same phenotype. nothing can be said about the sex determination. And if both constitutions are of the same sex or if one of them is a sexually non-functional intersex, it would be impossible for the tetraploids to maintain themselves by sexual reproduction. However, a somatic doubling of the diploid heterozygous sex would give tetraploids of the constitution XXYY, which would probably be of the same sex as XY. Tetraploid aspens originating by somatic doubling have been experimentally produced by means of colchicine treatment of germinating seeds (unpubl.).

IX. DISCUSSION.

The amentiferous plants are in general considered to be primitive and of ancient origin. *Salicaceæ*, comprising *Salix* and *Populus*, is the most primitive family. In this group the x-number is 19. Other members of *Amentiferæ* have x-numbers 14 (*Alnus, Betula*), 8 (*Carpinus*). 12 (*Fagus, Quercus, Castanea*), 16 (*Juglans*) (TISCHLER, 1926, 1931, 1936, 1938). The x-number 19 in *Populus* and *Salix* cannot be considered to be a primary one. In *Pyrus* the x-number 17 is considered to be derived from a primary x-number of 7. This conclusion is drawn mainly from the evidence of »secondary pairing» (DARLINGTON and MOFFETT, 1930). Also in other species secondary pairing has been used as evidence of genome complexity (cf. DARLINGTON, 1937). In *Populus* and *Salix* secondary pairing has never been reported. MOFFETT's view that plants with 34 + 7 chromosomes in the progeny of crosses between diploid and triploid apples are more viable than other individuals (MOFFETT, 1931) has been shown to be wrong by DERMEN (1936;

cf. WANSCHER, 1939). But in spite of this the assumption of a genome complexity affords a possibility of explaining the high viability of aneuploid apples. In this respect *Populus tremula* behaves intermediately between *Pyrus malus* and other species. Thus it seems as if the complex genome of *Populus* has lost almost all features of its origin, and on the whole behaves as a primary one. The only evidence is the rather high viability of aneuploids. This may indicate a more ancient origin than that of *Pyrus,* in agreement with the view of most taxonomists, that *Populus* belongs to an old group of plants. Both *Populus* and *Salix* are dioecious. Heterochromosomes have been reported for several species, but at least as far as *P. tremula* is concerned this report is doubtful. Polyploidy has been of a very different importance in the two genera. In *Salix* polyploidy is common, diploid, tetraploid and hexaploid species are reported (BLACKBURN and HARRISON, 1924). In *Populus* no polyploid species is known for certain. In spite of this, triploid clones of *P. tremula* are fairly common. In *P. alba* and *P. canescens* — probably a hybrid between *P. tremula* and *P. alba* — triploids have been reported (PETO, 1938). These triploids have probably originated due to unreduced gametes (MÜNTZING, 1936; PETO, 1938). The occurrence of extremely large — presumably unreduced — pollen grains by diploid *Populus* species has been found by PETO (1938). The triploids, too, produce »giant» pollen grains, as shown by MÜNTZING (1936) and PETO (1938). In artificial crossings between a diploid female and a triploid male *P. tremula* about 3 % of the offspring were found to be tetraploids. Thus there is a great possibility of intraspecific tetraploids arising in nature. Interspecific hybrids within *Populus* are common in nature (HOUTZAGERS, 1937). One diploid hybrid has been shown to produce large pollen grains and one hybrid to be a triploid, producing large pollen grains (PETO, 1938). For that reason there is a great possibility also of interspecific polyploids occurring in nature. Thus the lack of tetraploid and higher polyploid *Populus* species and races must be attributed to special factors. The fact may be that eventually arising tetraploids for some reason or other cannot survive in the competition in nature. Another factor well worth mentioning is the dioecy. Of course a single tetraploid, if purely dioecious, cannot maintain itself by sexual reproduction. The condition for this must be that two tetraploids of opposite sexes arise in the neighbourhood of each other. Such events may be statistically rare, but, no doubt, the natural population has in the course of time been large enough to create them. And in the related genus *Salix* polyploidy

is common in spite of dioecy. Furthermore, some *Populus* species —
for instance, *P. tremula* — are capable of vegetative propagation in
nature by means of suckers, and for that reason these species have an
ability of migrating over large areas. Thus the questions concerning
the absent natural polyploidy in *Populus* are at present impossible to
answer. Further experiments may perhaps provide the clue.

SUMMARY.

1. Some new triploid *Populus tremula* clones in Sweden have been
found. Altogether nine triploid clones are known.

2. Data are given, showing the connection between leaf size, lengths
of stomata and 2n-numbers. It is pointed out that large leaves are
always correlated to long stomata, but not always to triploidy.

3. The pollen properties of the triploids, compared with those of
the diploids, have been investigated. The triploids have larger pollen
grains and poorer pollen than the diploids, but the diploid and triploid
variation overlaps.

4. Among the triploids pure males and pure females exist. The
type of sex determination is discussed.

5. Meiosis of diploid male aspens has been studied. In general,
diploid meiosis is quite regular. Four diploid clones out of sixteen have
shown a varying frequency of univalents. The behaviour of the uni-
valents is described.

6. Meiosis in triploids follows the general course described by
MÜNTZING (1936). Some further results are added.

7. Crosses between diploid and triploid aspens have been produced
in both directions.

8. The 2n-numbers of 617 offspring plants are given. Intermediate
2n-numbers between 2x and 3x are represented in a high frequency,
but the distribution is not binomial. Reciprocal crosses give the same
result. In the progeny 18 $\pm$ 4x-plants were found. The occurrence
of tetraploids is ·attributed to the formation of unreduced gametes in
the triploid parent.

9. The deviation of the progeny from the binomial distribution is
attributed to numerically selective elimination. It has been shown that
this elimination involves gametes, zygotes and embryos as well as seed-
lings and young plants. The elimination of gametes as well as zygotes
and embryos is approximately identical in reciprocal crosses.

10. Measurements of stomata and plant-height of the progeny are

given.   No correlation between stomata-length and 2n-number within the 2x—3x variation was found, but on an average the aneuploids had longer stomata than the diploids.   The tetraploids had longer stomata than both aneuploids and diploids.   Plant-height is very variable among the aneuploids.   The aneuploids are poorer than the diploids.   No conclusions can be drawn as to the vigour of the tetraploids, because of the fact that photoperiodic races exist within *P. tremula* (SYLVÉN, unpubl.), and the tetraploids are occasionally produced by crosses which do not give progenies, adapted to the light climate of the experimental station.

11.   The possible sex of the tetraploids is discussed.

12.   The x-number and the lack of polyploidy within the genus *Populus* is discussed.

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CONTENTS.

THE CYTOLOGY OF ALLIUM AMPLECTENS AND THE OCCURRENCE IN NATURE OF ITS ASYNAPSIS

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INTRODUCTION.

MOST of the *Allium* species occurring in the Pacific Coast region of North America are endemic and closely related. They form a morphological type readily distinguishable from the *Allium* species of the Old World, which exhibit a decidedly greater diversity. The relative uniformity of the American *Allium* species is reflected in their chromosome conditions: all West-American species so far studied have the basic number 7, while the Eur-Asiatic species usually have the number 8, although the numbers 7 and 9 are also found among them. Unfortunately the taxonomy of the genus *Allium* is not sufficiently known to allow any conclusions concerning any possible relationship of the American species to any certain group of the Eur-Asiatic species. A study along these lines should give important results, among others. in phylogenetic and plant-geographical respects.

In order to prepare the ground for a joint attack on the taxonomy, cytology and species formation of the genus *Allium*, I have for some years examined different small groups of species from a cytological and in certain especially favourable cases also phylogenetic view-point. *Allium* must be regarded as an ideal material for such studies. About 300 species are distributed all over the northern hemisphere. They are characterized by remarkably well-defined and constant cytological features. The idiograms of the species may be studied with an almost schematic clearness during the first microspore mitosis. Several species may be recognized only by the appearance of their satellited chromosome. And cytologically observable variation, both structural and numerical, very often gives the key to the type formation within related groups of taxonomic units.

In the present paper I take up for examination a West-American species, *Allium amplectens* TORR. This species is of especially great interest because of a genetic abnormality of meiosis, present in certain

forms of the species. Such gene-controlled deviations from the normal course of meiosis are by no means rare in *Allium*. On several earlier occasions I directed attention to the formation of dyad pollen due to failure of the second meiotic division, an abnormality now and then found in most *Allium* species. In *Allium amplectens*, on the other hand, asynapsis leads to the complete failure of the first meiotic division, a state of things not previously encountered in *Allium*.

The outlines of this asynapsis process have already been published (LEVAN, 1938). The process differs in certain fundamental respects from all cases of asynapsis previously described. Its main characteristic is its completeness, which is associated with great regularity in the asynaptic meiosis. In fact, the asynaptic meiosis takes place with fewer disturbances than meiosis in normal synaptic plants of the same species, which suffer from the usual disturbances due to autopoly-ploidy. The pollen formed after the asynaptic meiosis has a regular shape, deviating from normal *Allium* pollen. This permits a rapid diagnosing of asynapsis even in dried material. On account of this very favourable condition, the investigation of the cytology of the species could be completed by a plant-geographical study.

In the first part of this paper the cytology of normal and asynaptic *Allium amplectens* will be treated. The material of this part consists of a few forms of the species which I brought home from California. They turned out to include samples of both types of meiosis. The second part of the paper is based on herbarium material from different parts of the distribution area of the species. The asynapsis, studied in detail in the first part, can now be placed in its plant-geographical connexion.

California Academy of Science, San Francisco, and Stanford University lent me dried material of the species. Dr. CARL W. SHARSMITH, Curator of the herbarium, State College of Washington, Pullman, and the Librarian, Bureau of Plant Industry, Department of Agriculture, Washington, D. C., helped me in various ways. Miss ANNA NORD-STRAND, Hilleshög, has given me valuable technical assistance.

I. CYTOLOGICAL STUDIES.

1. MATERIAL AND METHODS.

The material for the cytological study of *Allium amplectens* was collected on an excursion on the 13th May, 1937, in Santa Clara County, California. Six different forms were collected. They are listed below together with Dr. KECK's description of the localities:

7043. (KECK No. 4531). Mt. Hamilton — San Antonio Valley road, at Isabel Creek, just east of crossing, in creeklet. Bulbs in mud. No plants in neighbouring slopes.

7042. (KECK No. 4537). Arroyo Bayo, 5 miles east of Isabel Creek, Mt. Hamilton range. In alluvial flats and in creek bed.

7048. (KECK No. 4540). Arroyo Bayo, 6,8 miles south-east of Isabel Creek, in moist creek bed.

7049—7051. (KECK Nos. 4549—4551). Beauregard Creek, 14,2 miles from crossing of Isabel Creek (31 miles from Livermore). In dry soil.

Among these forms, 7043, 7049 and 7051 turned out to be asynaptic and triploid, while the rest had normal meiosis and were tetraploid. Another normal tetraploid of *Allium amplectens* (my No. 657) was investigated earlier (LEVAN, 1931). It had been procured from the Botanic Garden of Copenhagen.

Since these forms of *Allium amplectens* cytologically represent two types, the normal and the asynaptic, no distinction will subsequently be made between the different forms within each type. I wish to point out, however, that numerous slides from different fixations have been studied of all forms. They all showed good agreement in their main cytological features. It may therefore be concluded that the asynapsis is not due to modificative influences or mere chance occurrences.

The general appearance of two of the investigated forms is seen in Fig. 1 *b—c*, which represent one triploid asynaptic form and one tetraploid normal form. Fig. 1 *a* is a related diploid species, *Allium*

serratum, collected in the same region as *Allium amplectens*. A close picture of the *amplectens* flowers is shown in Fig. 10.

The cytological methods employed are the same as those previously described (LEVAN, 1932, 1936), viz. meiosis fixed in whole flower buds in NAVASHIN or in smears in BENDA—GEITLER, pollen fixed in smears in BOUIN—ALLEN. The drawings were made with the aid of the following lens system: ZEISS apochromatic objective H 120 × 30 or in general


Fig. 1 *a*: *Allium serratum*, *b*: *Allium amplectens*, triploid form, *c*: ditto, tetraploid form.

views × 15, which on drawing gives a magnification of 6500 and 3250 times respectively.

2. SOMATIC CHROMOSOMES.

The basic number of *Allium amplectens* is 7, as already mentioned. Diploid forms with the somatic number 14 occur probably in nature (see Chapter II: 3), but no such forms were found among the living material available for cytological study. The forms cultivated by me

were triploids and tetraploids with the somatic numbers 21 and 28 respectively.

All these forms agree in their chromosome morphology. The chromosomes are all of one type, two-armed with medially or sub-medially located centromeres (Fig. 7). Satellites are entirely missing. And since even very small satellites are easily seen in the pollen mitoses of related species, for instance *Allium serratum* (Fig. 9 *a*, *b*), it is highly probable that *Allium amplectens* is really devoid of satellites. The very peculiar nucleolar conditions present in *Allium amplectens* (cf. Chapter I: 6) are presumably connected with this absence of satellites.

The chromosome size is very large, even compared with other *Allium* species. The longest chromosomes of the idiogram have during the pollen metaphase a length of up to 15 μ, while their breadth in the neighbourhood of the centromeric constriction is somewhat less than 1 μ. It has been previously shown (LEVAN, 1935 a) among other *Allium* species that members of the 7-series have on an average longer chromosomes than 8-chromosome species. While the average length of the former was estimated at 13 μ, the latter had a length of about 9 μ. The chromosomes of *Allium amplectens* are exceeded in size only by another American species, viz. *Allium (Nothoscordum) fragrans*.

3. MEIOSIS OF THE NORMAL TYPE.

Meiosis of all the tetraploids investigated takes place without any deviations from the normal course of meiosis already studied in several autotetraploid *Allium* species. Thus it agrees completely with meiosis of the only other known tetraploid American species, *Allium validum*. All stages of meiosis could be studied with great clearness. Already at pachytene very evident quadrivalents were observed (Fig. 2 *a*—*c*). The pairing was very good, the unpaired threads frequently met with could often be seen to be due to exchanges of partners within the quadrivalents. Due to the large size of the chromosomes and chromomeres the differences in shape and size of individual chromomeres could easily be demonstrated, in the same manner as BELLING did (1928) in *Lilium*. It was found that the paired chromomeres of homologous threads were usually identical in shape. Very striking exceptions from this rule were not so seldom met with, however, and it was noticed that within regions where dissimilarities in appearance between the paired chromomeres were present, the pairing seemed to be loosened, loops of unpaired threads being formed. An instance of this is pictured in Fig. 2 *d*.

Both the shape and number of chromomeres within the pictured region are quite different in the two threads.

In diplotene normal chiasmata are seen (Fig 2 e—g), some chiasmata holding the chromosomes together into quadrivalents. Other quadrivalents separate, due to lack of chiasmata, into 2 bivalents or one
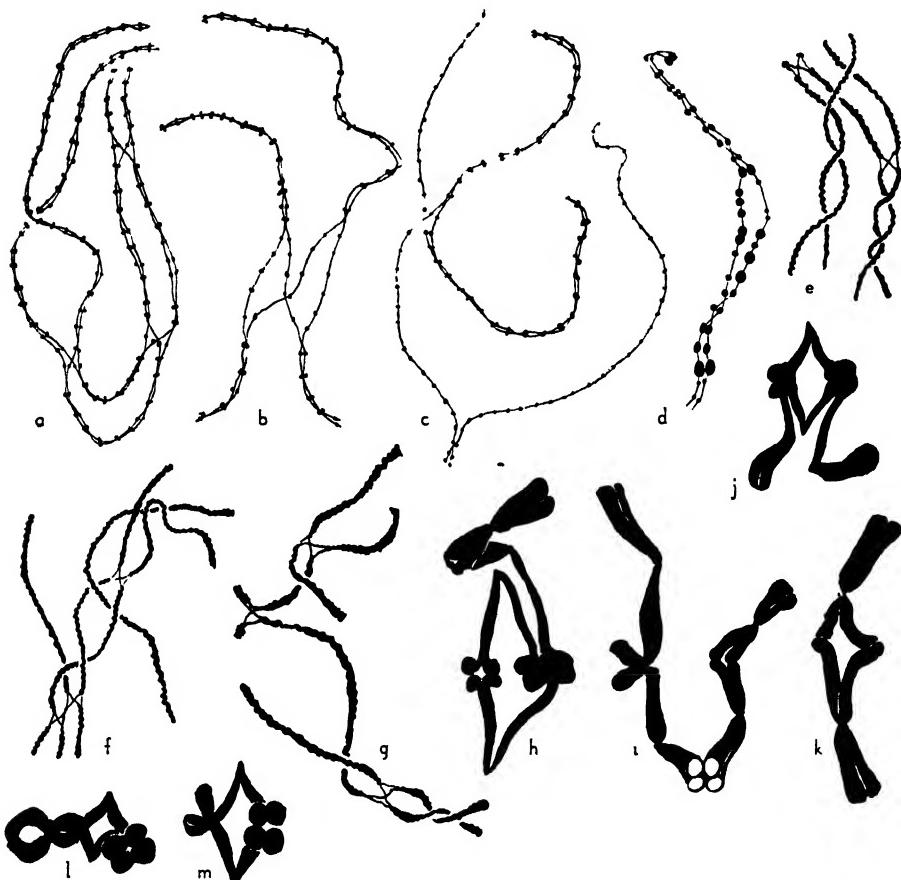

Fig. 2. Meiosis of the normal type, a—d: pachytene, e - g: diplotene, h--m: meta-
phase I. — × 3250 (except d, which is a free hand drawing).

trivalent and one univalent (as in Fig. 2 e and f). At diakinesis and metaphase I the chromosomes assume the typical meiotic appearance. It is now possible to analyse whole cells, and it may then be seen that one or two quadrivalents occur in almost every cell. In single cells all the chromosomes may remain associated into 7 quadrivalents. Fig. 2 h—j shows a couple of the most common types of quadrivalents.

Chains of four were commonest, and single rings and frying-pan quadrivalents occurred somewhat more rarely. The bivalents (Fig. 2 k—m) might have 1—5 chiasmata, commonly 1—2.

At the first anaphase very often a number of univalents were present. In one slide the following frequency of univalents was counted:

Number of univalents:	0	1	2	3	4	Total	Med.
Number of cases:	4	7	12	6	5	34	2,0

The univalents always formed a secondary equatorial plate and were eventually divided in the first division. This causes a certain disturbance of the first division, resulting in the formation of micronuclei in the interkinesis. The following number of micronuclei was determined in one slide:

Number of micronuclei:	0	1	2	3	4	5	Total	Med.
Number of cases:	6	10	14	8	4	1	43	1,9

These irregularities of meiosis give rise to a somewhat varying number of chromosomes in the pollen grains.

4. MEIOSIS OF THE ASYNAPTIC TYPE.

The zygotene pairing of the asynaptic type takes place absolutely normally in the manner previously described in synaptic triploid *Allium* species, e. g. *Allium Schoenoprasum* and *carinatum*. Probably 7 trivalents are formed in most cells. Trivalents which I was able to analyse at pachytene usually had 2 pairing blocks (Fig. 3 a—d). I wish especially to emphasize that the chromomere paring within the frequently very clearly visible pachytene configurations was, as far as could be seen, of the same strong and intimate nature as is found in the synaptic types. Thus, it is impossible at this stage to distinguish the asynaptic type from normal types with complete pairing.

Already at early diplotene, however, a striking difference from the normal type is observed. All points of crossings within the trivalents now turn out to consist of mere overlappings and no real chiasmata at all are found. It is evident that the formation of chiasmata has failed

in some way or other. That the three chromosomes of the trivalents are still placed together (Fig. 3 *e—j*) is due to their relational spiralis-
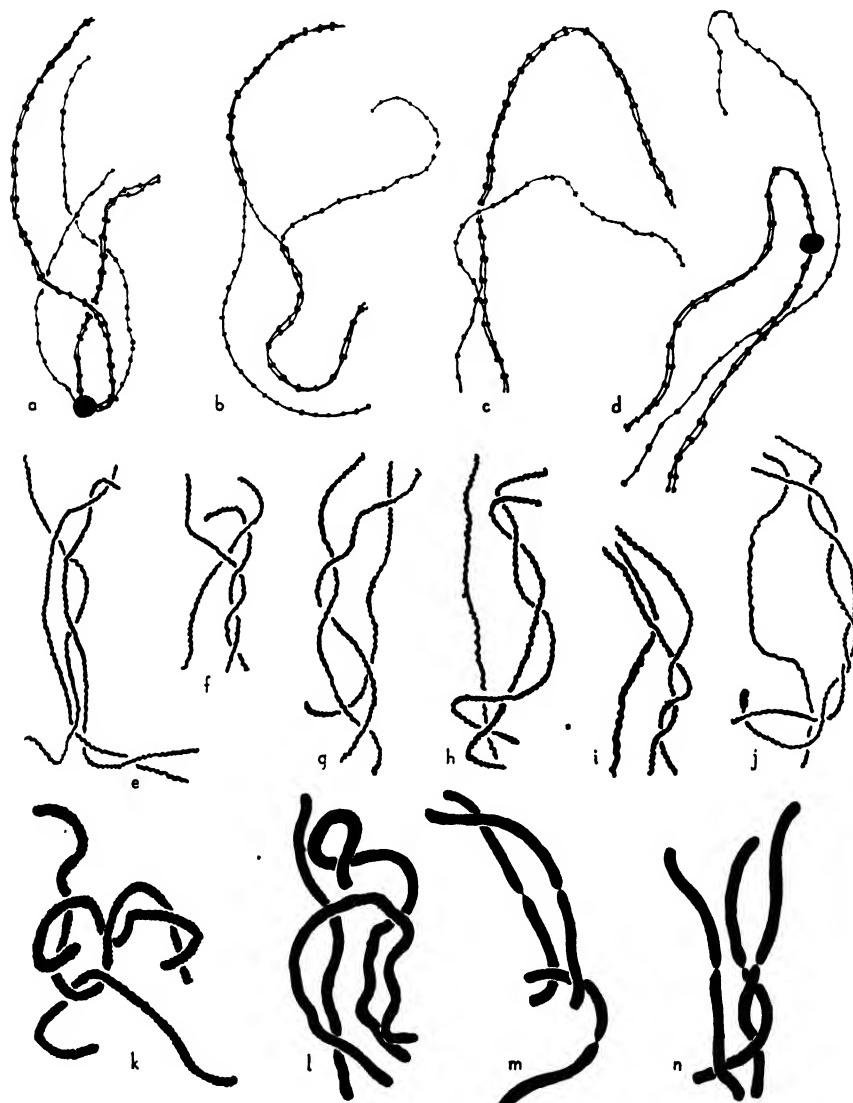

Fig. 3. Meiosis of the asynaptic type, *a—d*: pachytene, *e—j*: diplotene, *k—n*: dia-kinesis. — × 3250.

ation. Because of this it is often possible, even at late diplotene and diakinesis, to recognize the original pairing blocks (Fig. 3 *k, l*).

At the same time as the contraction of the chromosomes increases,

the relational spirals uncoil and numerous free univalents begin to appear. Their position, however, indicates for a long time which chromosomes originate from the same trivalent. At diakinesis (Fig. 3 *m—n*. Fig. 4 *a—c*) the centromeric region of the chromosomes may be clearly discerned as a constriction medially located in all chromosomes. The chromosomes gradually acquire a more somatic metaphase appearance,
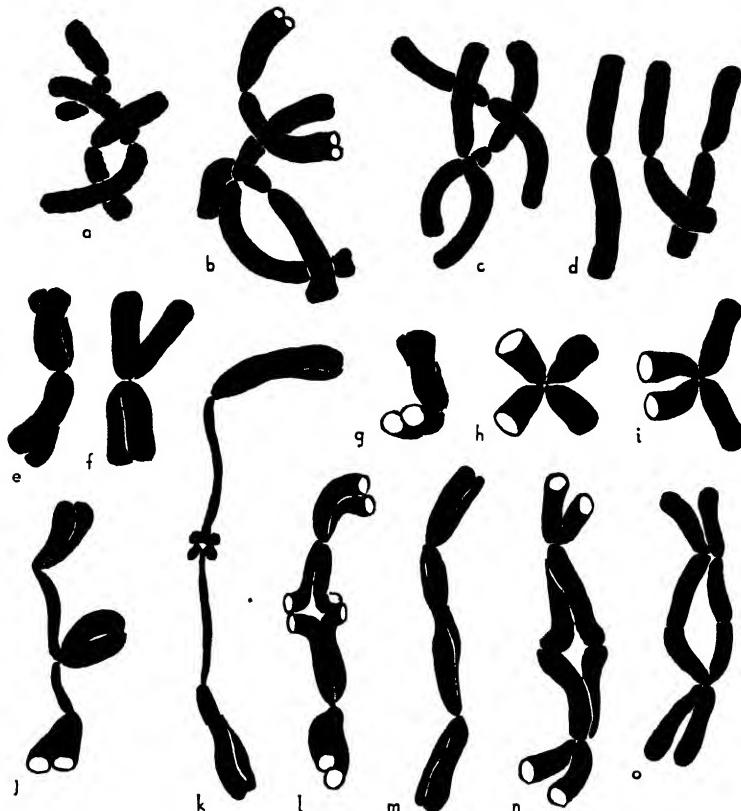

Fig. 4. Meiosis of the asynaptic type, *a—c*: diakinesis, *d*: metaphase I, *e —i*: the splitting of the metaphase I univalents, *j- -o*: bivalents of the asynaptic type, seen in polar view. — × 3250.

and when the nuclear membrane disappears they resemble ordinary root chromosomes, even if their contraction is somewhat stronger. The split between the daughter chromatids, which was quite apparent during diplotene and diakinesis, has by now become invisible (Fig. 4 *d*).

Here ceases, however, the resemblance to somatic chromosomes, in which, at this stage, the centromeres would very soon have divided. In the asynaptic metaphase, on the other hand, the centromeres

remain undivided.   The chromosomes are arranged with their longitudi-
nal direction extended in the polar plane and proceed towards the
equator (Fig. 6 *b*).   There is usually not space enough for all 21 uni-
valents in one equatorial plate, so 4 or 5 chromosomes form accessory
plates on one or both sides of the equator.   During this process the
daughter chromatids begin to fall apart.   Since the centromeres are
still single, however, each chromosome assumes a more or less typical
cross-shape (Fig. 4 *e—i*, Fig. 6 *c*).
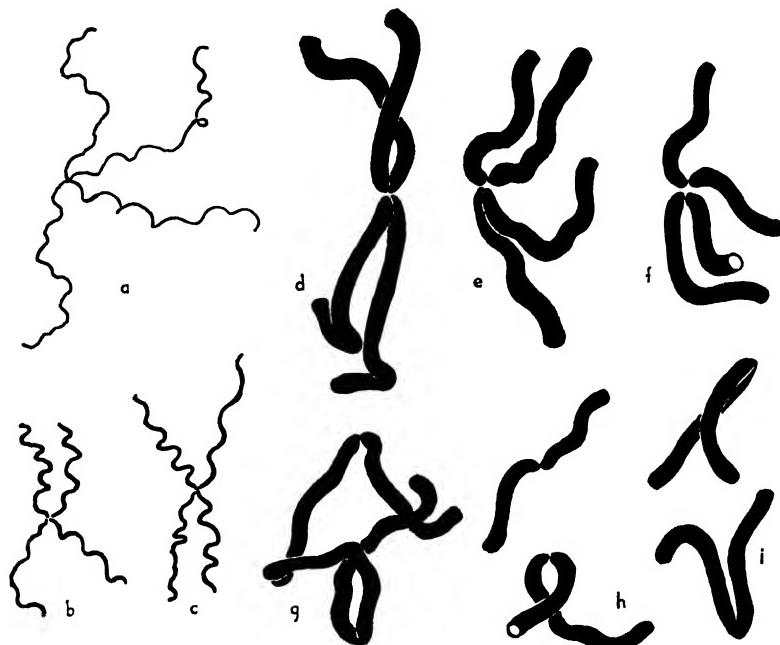
Fig. 5.   Meiosis of the asynaptic type, *a -c*: univalents at interkinesis, *d—f*: second
metaphase, *g—i*: second anaphase. — ✕ 3250.

A great number of slides of this stage was examined, and I then
found that a chiasma could really be formed now and then.   The
frequency of these chiasmata varied and was always very low.   In most
slides one chiasma could be found in some 500 cells, in other slides one
chiasma in 100 cells and in one slide even 4 chiasmata in 100 cells.
These chiasmata gave rise to bivalents and in 2 observed cases to tri-
valents.   Considering the extremely low frequency of chiasmata it is
impossible that a random distribution of chiasmata should bring about
any formation of trivalents.   The 2 chiasmata within the observed tri-
valents must consequently be due to the same cause, presumably a

change in the precocity conditions within the three chromosomes of the pachytene trivalent.

The studied bivalents, some of which are pictured in Fig. 4 *j—o*, all had one chiasma each. This chiasma was in most cases wholly terminalised, but in a few cases an interstitial position of the chiasma was observed. In one case it was situated about half-ways out on the chromosome arms (Fig. 4 *j*). The fact that the chiasmata of the bivalents were usually terminal suggests that the repulsion between the centromeres of the bivalents was of normal effect. Also the shape of the bivalents indicated the same thing: the portion of the bivalent lying between the chiasma and the centromeres was very often extended into a narrow thread (Fig. 4 *k*).

This condition is perhaps somewhat surprising, since the orientation of the bivalents in relation to the poles was always quite different from the normal bivalent orientation. In no case were the centromeres of the bivalents observed to be co-orientated, on the contrary, they were always situated within the equatorial plate at the same level as the centromeres of the univalents. And it could happen that the arm connecting the two centromeres of the bivalent ran right through the whole plate.

This condition has a certain significance for the understanding of the mechanics of the centromeres. Evidently it is not the chiasma formation *per se* that causes the characteristic orientation of the meiotic bivalents, rather it must be some quality within the centromeres themselves. It may be concluded in accordance with DARLINGTON (1937) that a polarisation of the centromeres brings about a predisposition to auto-orientation, while a lacking of polarisation causes co-orientation. And this polarisation has apparently nothing to do with the presence or absence of chiasmata. In the solitary bivalents of the asynaptic meiosis the orientation is the same as that found earlier in the so-called somatic bivalents, originated by segmental interchange between different somatic chromosomes. The further bearing of this behaviour of the centromeres on the interpretation of the asynapsis of *Allium amplectens* will be discussed in the final chapter.

After some time the chromosomes begin to exhibit a decreased stainability and an evident despiralisation, in fact, they begin to show telophase characters. Sometimes it may appear as if the centromeres had divided, the daughter chromatids lying almost parallelly. This is, however, a false appearance due to the fact that the region of the chromosomes close to the centromeres loses its stain somewhat earlier

than the rest of the chromosomes. The subsequent stages show clearly
that the centromeres remain undivided all through interkinesis. The
nuclear membrane begins to develop, sometimes separately around each
chromosome, usually, however, around the whole chromosome group.
The chromosomes of the periphery very often form nuclear lobes of their
own, communicating with the large central part of the nucleus. The
large single telophase nucleus formed in each pollen mother cell has at
first a peculiar disciform shape, due to the fact that the chromosomes
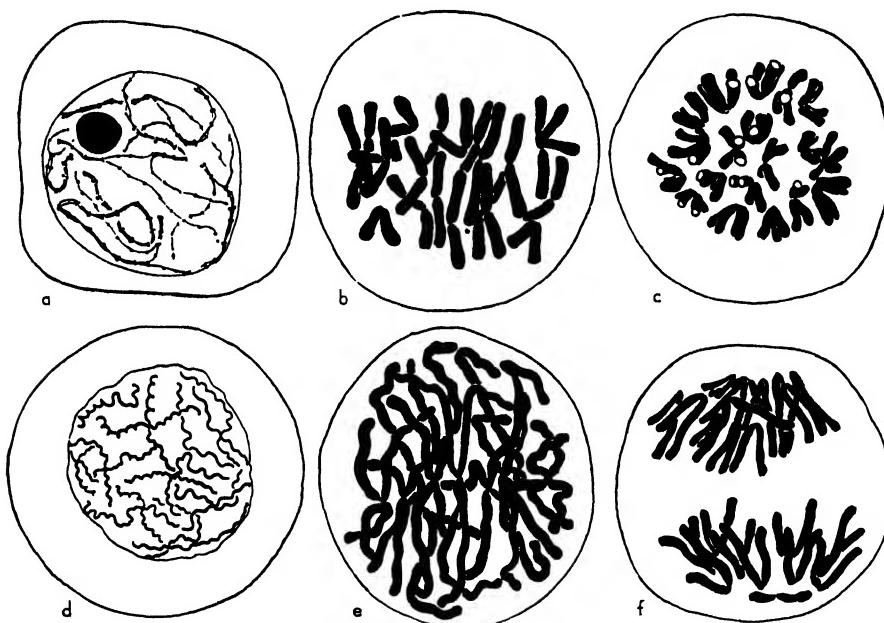
Fig. 6. Meiosis of the asynaptic type, the general outlines, *a*: pachytene, *b*: meta-
phase I, side view, *c*: a somewhat later stage of metaphase I, polar view, *d*: inter-
kinesis, *e*: metaphase II, *f*: anaphase II. — × 1400.

were overtaken by the telophase, while the centrosomes were still
keeping them strictly arranged on the equatorial plate.

Thus a uninuclear interkinesis stage is formed with complete
regularity (Fig. 6 *d*), a kind of restitution nucleus containing the total
of the first meiotic chromosome quantity. Even in these cases, where
from the beginning numerous micronuclei were formed, they later on
fuse into one single nucleus. And if in exceptional cases more than
one interkinesis nucleus are maintained until the beginning of the
homeotypic division, they will never form separate spindles in this
division, they will instead be included in the main spindle.

During the interkinesis stage a clear spiral structure of the chromosomes may be seen (Fig. 5 *a—c*). When the nuclear membrane disappears at the onset of the second division, the centromeres are already from the beginning arranged in the equatorial plane, while the chromosome arms are folded towards the poles (Fig. 5 *d—i*). The chromosomes now fill up the cell in quite a different manner from that at the first metaphase (Fig. 6 *e*).

Now the centromeres are finally divided and a normal anaphase starts (Fig. 6 *f*). This proceeds normally and the result is pollen dyads with somatic chromosome number. The regularity of this meiosis type is proved very clearly by the conditions of the pollen described in detail in the next chapter. All investigated pollen grains have exactly 21 chromosomes. Thus, the asynaptic meiosis is a rather unique way in which a triploid species has succeeded in acquiring the ability of forming pollen grains with balanced chromosome conditions.

5. THE MITOSIS OF THE POLLEN GRAIN.

a. *The normal type.* — The pollen tetrads of the synaptic forms have the normal *Allium* appearance. The pollen grains are crescent in shape (Fig. 7 *a*, Fig. 8 *a*). As expected, 14 is the most common chromosome number of the pollen, but the number 13 is also found fairly often. In a couple of slides the following frequency of chromosome numbers was noted:

Number of chromosomes:	12	13	14	15	Total	Med.
Number of cases:..................	2	15	24	3	44	13,6

b. *The asynaptic type.* — The asynaptic dyad pollen is semispherical in shape (Fig. 8 *b*) and resembles completely the pollen grains originated by monokinetic meiosis (LEVAN, 1935 b, Fig. 46). The first mitosis of the asynaptic pollen grains occurs quite regularly. The chromosome number is 21 (Fig. 7 *b*). The spindle is asymmetric and at anaphase the larger anaphase group goes up distally into the pollen cupola, while the more condensed group is pressed towards the proximal, plane pollen wall. The former group forms the vegetative nucleus and the latter the generative nucleus. The asynaptic pollen shows the same germinating power on agar as normal pollen. The

division of the generative nucleus occurs in the pollen tube about 24 hours after the onset of germination.

The constancy in chromosome number of the asynaptic pollen is in fact rather remarkable. All 36 metaphase plates of one slide, the chromosome number of which could be determined with absolute certainty, contained exactly 21 chromosomes. This furnishes better than anything else a picture of the regularity with which the asynaptic meiosis takes place. In this respect *Allium amplectens* is different from previously described cases of asynapsis, where the simultaneous presence of univalents and bivalents upsets the balance of meiosis.

Among the asynaptic dyad pollen there is always a very low but
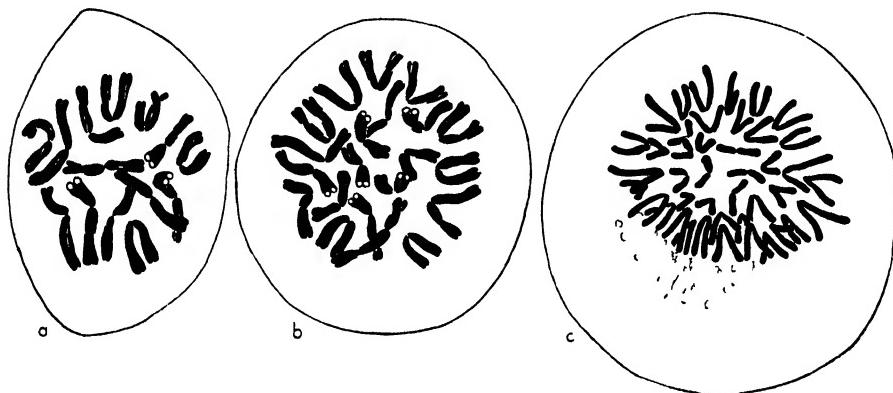
Fig. 7. The first pollen mitosis, *a*: normal tetrad pollen ($n = 14$), *b*: asynaptic dyad pollen ($n = 21$), *c*: asynaptic monad pollen ($n = 42$), *a*, *b*: metaphase, *c*: anaphase. — $\times 1200$.

regular percentage of monad pollen. An idea of its frequency may be had from the fact that in one slide ($= 5/2$ anthers) 30 monads were present. The frequency is probably less than 1 $^{0}/_{00}$. The mode of origin of the monad pollen grains could not be directly observed, owing to their extreme rarity. I think, however, that there is little doubt that they originate by monokinetic meiosis, which is a very common process in *Allium*. In this case, where also the first division is omitted, it should perhaps more appropriately be called akinetic meiosis.

The monads are spherical in shape and their diameter is decidedly larger than in the dyad pollen. One monad pollen grain can be seen in the microphoto, Fig. 8 *b*, below the centre at about 7 o'clock. The increase in cell diameter is of interest. Evidently the plane, proximal pollen wall of the dyads puts a limit to their further growth in size, whereas the spherical monads may continue growing still further.

The chromosome number of the monads has been directly counted only in a few cases. In a couple of anaphases, one of which is pictured in Fig. 7 c, there were 42 chromosomes present, i e the double somatic number. Although those moments of asymmetry, due to the tetrad division, are absent in the monads, their pollen mitosis is markedly asymmetric One anaphase group is spread out in the centre of the cell, developing into the vegetative nucleus, the other group is pressed against the wall into the flattened, lentiform, generative nucleus The situation
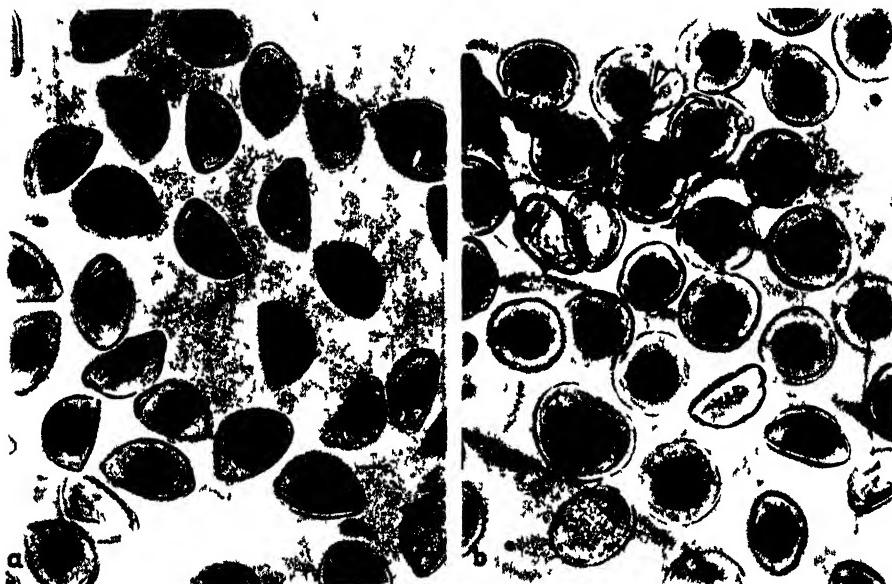

Fig 8 Microphoto of pollen grains of *a* normal type *b* asynaptic type ×300
Microphoto OTTO MARISSON

is exactly similar to that seen in the artificially produced monad pollen (LEVAN, 1939) The monad pollen, too, may be easily germinated on agar

It should be observed that the 21-chromosome pollen grains of the asynaptic form all contain the same genic quantity as somatic cells No genotypic variation occurs between different pollen grains, a variation generally present elsewhere among the gametes of triploid forms. It is therefore possible here to make a comparison between the purely modificative chromosome variation between different pollen grains and the variation due to genetic segregation. The genic variation is especially pronounced in plants which are hybrids between

types with different chromosome size. This condition is seen very plainly in the hybrid *Allium Cepa* × *fistulosum*. Other instances of this are recorded by UPCOTT (1939). She emphasizes that the difference in size is due to a varying spiralisation »owing to the direct action of the individual genotype». It must be said, however, that even in the pollen of the asynaptic *Allium amplectens* there occurred a greater variation in chromosome contraction between different pollen grains than is generally seen in the root tissue of *Allium*. No doubt, however, the variation was somewhat less than in the pollen of synaptic triploids and hybrids.

6. THE NUCLEOLI.

Allium amplectens is, compared with most other *Allium* species, characterized by deviating nucleolar conditions. In most *Allium* species
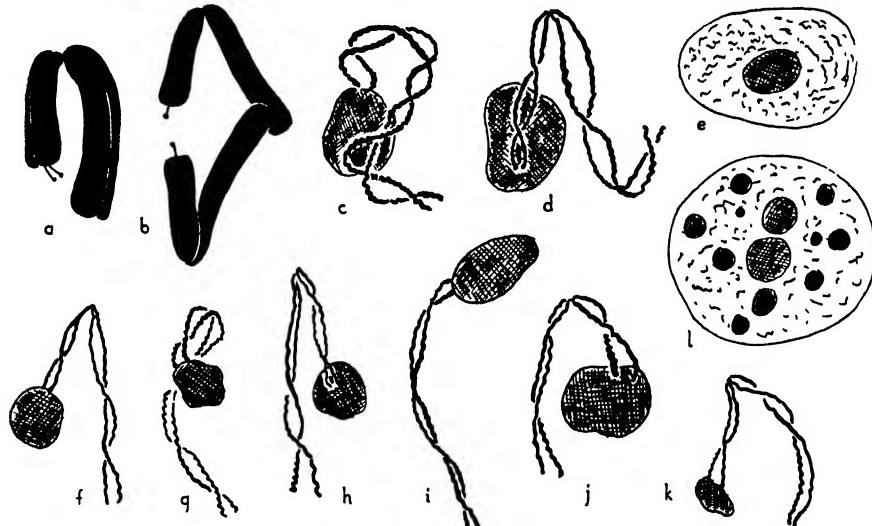
Fig. 9. Nucleolar conditions of the pollen grains, *a—e*: *Allium serratum*, *f l*: *Allium amplectens*, *a*: the s_1 chromosome at metaphase, *b*: at anaphase, *c—d*: 2 instances of the nucleolar chromosome, *e*: vegetative resting nucleus, *f—k*: 6 nucleolar chromosomes from one nucleus, *l*: vegetative resting nucleus. — *a—d, f— k*: × 3250. *e, l*: × 1600.

there is present one satellited chromosome (the »s_1») in each genome, and the nucleolus is formed at the achromatic portion of this s_1. Thus diploid species usually develop two nucleoli in somatic cells and one nucleolus in the pollen.

As mentioned above, no s_1 could be identified in *Allium amplectens,* and its nucleolar conditions are irregular. In recently divided somatic cells a great many nucleoli are formed. Systematic counts of the

nucleoli could not be made, since the conditions in somatic tissues are rather irregular and not so clear unless special stainings are used, but 10 or 12 nucleoli in each cell are very often present. If the number of nucleoli is great, they are tiny in size, if one or two decidedly larger nucleoli are present, the number of nucleoli is less. Thus it is clear that the nucleoli fuse.

In the archesporium and in very young pollen mother cells there are many nucleoli (about 4—10). But in the prophase stages of meiosis, pachytene and diplotene, not more than one nucleolus is seen. It is not very large and cannot be the result of the fusion of many nucleoli. It seems to be attached to the end of one paired pachytene chromosome.

The nucleoli are more clearly observable during the pollen development. In the prophase of the first pollen mitosis several nucleoli per cell are present. In the different types the following numbers were counted:

	Number of nucleoli:	1	2	3	4	5	6	7	Total	Med.
Number of cases	Synaptic type: ...	1	11	18	4	—	—		34	2,7
	Asynaptic type: ...	—	1	1	16	9	6	2	35	4,7

At late prophase it may be seen that the nucleoli are attached to individual chromosomes. In Fig. 9 *f—k* 6 such prophase chromosomes from the same cell of the asynaptic type are reproduced.

At late telophase of the pollen mitosis the nucleoli are present in great numbers. The following number was counted on one occasion:

	Number of nucleoli:	3	4	5	6	7	8	9	10	Total	Med.	Med. Ge-nome
Number of cases	Synaptic type: ...	5	20	8	3	—	–	—	—	36	4,3	2,1
	Asynaptic type: ...	—	—	1	5	12	9	4	2	33	7,2	2,4

Thus it may be concluded that the development of the nucleoli in *Allium amplectens* is not associated with one determined chromosome of the genome. 6 chromosomes of the synaptic pollen and 10 or even more of the asynaptic pollen are capable of producing nucleoli. Calculated on the number of chromosomes present between 2 and 3 chromosomes per basic genome may function as nucleolus carriers.

As a comparison I shall briefly mention the nucleolar conditions of the pollen of the diploid Californian species *Allium serratum*. Its basic chromosome set is of the same morphological appearance as the *amplectens* genome apart from the important difference that *Allium serratum* has a typical s_1 chromosome (Fig. 9 *a*, *b*). In the prophases
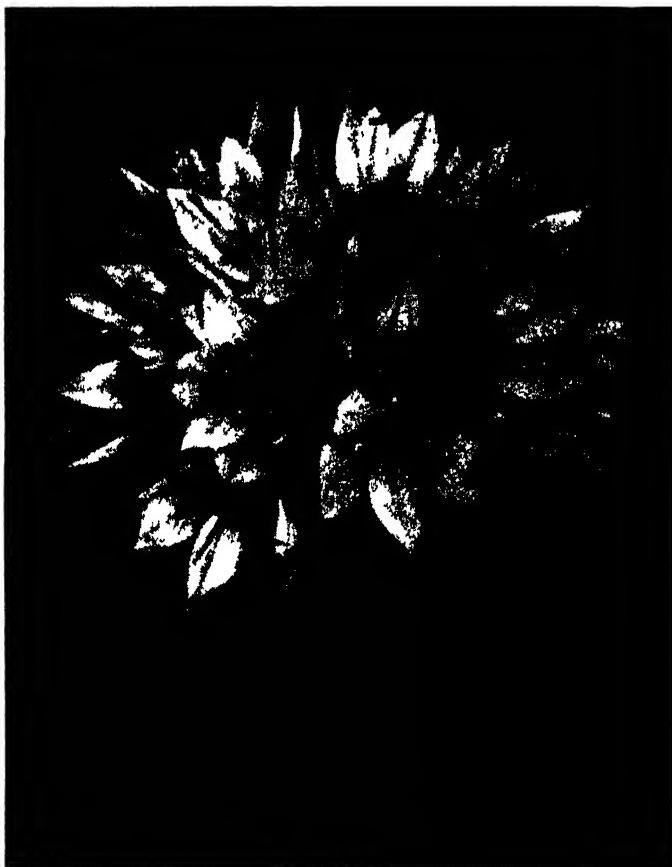
Fig. 10. Inflorescence of *Allium amplectens* showing the crests of the seed capsules.

and resting nuclei of the pollen there occurs regularly only one nucleolus (Fig. 9 *e*). This is formed distally on one chromosome, probably the s_1 (Fig. 9 *c—d*). I have not, however, been able to recognize the satellite in this stage, but its presence has been ascertained in several other *Allium* species.

·Between *Allium amplectens* and *serratum* there occurs a similar distinction to that MATSUURA (1938) observed between *Trillium* and

Paris. In the former, satellites are lacking and the nucleoli may be formed
on several chromosomes, in the latter case a typical satellited chromo-
some is present and functions as the satellite organizer. In agreement
with these findings MATSUURA divides the nucleolar chromosomes into
two main types: *1.* the ordinary interstitial type, where the nucleolus
is formed at the achromatic portion of the satellited chromosomes, and
2. the terminal type, where nucleoli may be formed at the end of several
chromosomes. In this latter case certain chromosomes often prevail
as nucleolar organizers and there is a kind of competition between the
different organizers. If for some reason or other the satellite-carrying
portion of the interstitial type is lost, a transition to the terminal type
may be brought about. Instances of this were found by McCLINTOCK
(1934) in *Zea.* Pollen grains lacking the nucleolus organizer of the
chromosome 6 developed many small nucleoli. In other cases micro-
nuclei devoid of the satellited chromosome have been seen to develop
nucleoli.

The »Ubiquität» of »SAT» chromosomes, argued by RESENDE
(1937), must, it is true, be considered to be valid in most cases. Certain
exceptions to the rule cannot, however, in my opinion, be denied. It
seems impossible to assume that in *Allium amplectens* so many satel-
lited chromosomes should have been overlooked as is indicated by the
number of nucleoli. Especially in the pollen mitoses the chromosomes
are so clear that even very small satellites should have been observed
in some of the hundreds of metaphase plates which have been analysed.
Thus it is very tempting to assume, in accordance with MATSUURA, that
the difference in nucleolar conditions between *Allium amplectens* and
neighbouring species is due to the lacking of satellited chromosomes in
Allium amplectens.

II. HERBARIUM STUDIES.

1. HISTORICAL SKETCH.

In order to study the occurrence in nature of the asynaptic meiosis
of *Allium amplectens* it was found necessary first to make a brief
examination of the taxonomy of the species and to determine the
geographical distribution of the species and of its main types. In
the present chapter a short survey will be given of the taxonomical
characters of the species as they have been described by different
authors.

The species *Allium amplectens* was described for the first time by

TORREY (1856) on material from Sonoma County, California. The name *amplectens* denotes the condition of the spathal leaves: »spatha e bracteis 2 orbiculatis concavis subacuminatis flores amplectentibus». This character is especially evident in the few-flowered forms. TORREY also concludes his description as follows: »Easily distinguished by the small few-flowered umbel, which is almost enclosed in the concave purple bracts».

KELLOGG (1861) described another type under the name of *Allium attenuifolium*, which according to current opinion belongs to *amplectens*. Describing material from Mt. Shasta (Siskiyou County, California), he emphazises the leaf characters of this form: »Leaves two, radical, stem sheathing at the broad membranous base, striate and channelled below, closely canaliculate above, very narrow and slenderly attenuated toward the filiform apex: margins slightly scabrous». KELLOGG's type is a stout many-flowered plant: »Umbel globose, many-flowered (50 to 80 or more)». He detects the important character of crests on the wall of the seed capsule: »Germ, color lively, pinkish capsule, turbinate, sub-three angled or three rounded cells, each cell slightly or obsoletely two-crested, central axis at the pistil depressed».

Allium amplectens afterwards reappears in GREY (1867) as *Allium occidentale*, while WOOD (1868) partly retains the name *amplectens* with a verbatim quotation of TORREY's diagnosis, partly takes up another name, *acuminatum* HOOK. *β gracile*, for *amplectens* material from Butte County, California. REGEL (1875) who generally treats the American species somewhat summarily, places *amplectens* as a synonym of *serratum* WATS.

GREENE (1894), in his »Manual of the Botany of the Region of San Francisco Bay», mentions KELLOGG's *Allium attenuifolium* and gives the important description of the structure of the bulb scales: »Bulbcoats white, with a delicate transversely sinuate or serrate reticulation, the vertical lines especially also minutely sinous». This character is easily recognizable and distinguishes the species from related species. JEPSON, in connection with *Allium attenuifolium* in GREENE's Flora, describes a new species, *Allium monospermum*, which is characterised by »capsule (by abortion) 1-celled, 1-seeded». The type-locality is Vaca Mts. in Solano County, California. In his own »Flora of California», however, JEPSON (1922) combines these two species under *Allium amplectens* TORR. Other modern Floras, for instance, ABRAMS (1923), JEPSON (1925) and MUNZ (1935), agree rather well as to the limitations of *Allium amplectens*.

Fig. 11. Different types of *Allium amplectens*, *a*: San Benito Co, *b*. Sonoma Co, *c*: Humboldt Co, *d*: Siskiyou Co.

From the above facts will be seen that *Allium amplectens* is a rather multiform species. As the different collections began more completely to represent the different regions of its distribution, it became gradually possible to obtain a clearer insight as to what characters were of taxonomic value and which varied from form to form. This will be seen very clearly from the following list, comprising some of the taxonomic authors' descriptions of the different organs of *Allium amplectens*.

Bulb. –– TORREY: large for the size of the plant; KELLOGG: (about *attenuifolium*) small, roundish, truncated; GREENE: bulb-coats white; JEPSON: bulb-coats commonly reddish; MUNZ: bulb-coats reddish to greyish.

Scape. –– TORREY: scapo flexuoso spithamaeo superne bifoliato; KELLOGG: scape terete, solid, glaucous, smooth (minutely speckled); GREENE: scape 10—18 in. leafy below.

Leaves. –– TORREY: foliis filiformibus, leaves scarcely a line wide, overtopping the scape; KELLOGG: leaves two; GREENE: leaves several, very long and slender; ABRAMS: leaves 2—4, shorter than the scape, narrow becoming convolute-filiform above the sheathing base.

Spathe. –– TORREY: e bracteis 2; KELLOGG: bracts 3, outermost larger, broad-ovate or oblong-ovate, short acuminate, sessile, membranous, 4- to 9-nerved or more; GREENE: bracts 2, short, abruptly pointed; JEPSON: bracts 3; ABRAMS and MUNZ: bracts 2.

Umbel. –– TORREY: pauci (3—6) flora; KELLOGG: many-flowered (50—80 or more); JEPSON: (about *monospermum*) pedicels 50—80 (in his floras 1922 and 1925) pedicels 25—35, umbel erect, usually dense; ABRAMS: umbel dense, almost capitate; MUNZ; pedicels 10--40, slender, 5—12 mm long.

Flower colour. - - KELLOGG: whitish (scarcely a pinkish tinge?), midrib of the petals pinkish; GREENE: white; JEPSON: (*monospermum*) pale-purplish, (1922, 1925) white or nearly so; ABRAMS: white or tinged with pink.

Perianth. –– TORREY: sepalis oblongis obtusiusculis; KELLOGG: petals ovoid-diamond-acute, slightly inflexed from the middle, the three inner a little narrower; GREENE: oblong-lanceolate-acuminate segments, 3—4 lines long; MUNZ: narrowly oblong-ovate, 6—8 mm long.

Stamens. –– TORREY: filamentis e basi lata submonadelpha subulatis; KELLOGG: filaments inserted at the base, subulate, white; JEPSON: (*monospermum*) filaments with broadly deltoid and connate bases.

KELLOGG: stamens as long as the perianth; JEPSON: perianth more

or less exceeding the stamens and style; ABRAMS: stamens scarcely shorter than the perianth; MUNZ: stamens shorter than the perianth.

Capsule. — TORREY: capsula trigastrica, apice depressa, loculis dispermis; KELLOGG: cells 2-seeded, rarely more than one perfected; WOOD: semine unico; JEPSON: (*monospermum*) capsule 1-celled, 1-seeded.

From the often discrepant data cited above a rather good picture may be had of the variation of the species. In the subsequent list of herbarium specimens this picture will perhaps be supplemented in a few respects.

2. HERBARIUM MATERIAL EXAMINED.

In the following list of herbarium specimens some morphological observations are given, especially if the types examined deviate in some way or other from the normal *amplectens* type, or if several specimens from the same region or similar type of locality show a decided agreement in general appearance, which has been interpreted as characteristic of a certain ecotype. The specimens are arranged geographically into different counties; see the map (Fig. 12). For each listed specimen the following data are given: locality, my cytological number (in *italics*), year of collection, collector, herbarium in which the specimen is kept (B = University of California, Berkeley; C = California Academy of Science, San Francisco; D = Dudley Herbarium, Stanford University).

CALIFORNIA.

1. San Diego Co. Cuyamaca, *35*, 1880, S. B. and W. F. PARISH, D; *36*, 1903, L. R. ABRAMS, D; *21*, *37*, 1928, I. L. WIGGINS, BD; *5*, 1932, M. E. JONES, B.

This, the most southern locality of the species, is isolated from the rest of its distribution area. All the studied specimens are of the same, very characteristic type: scapes erect, rather thick and stiff, umbels dense, capitate, pedicels short. All specimens have asynaptic pollen.

2. San Luis Obispo Co. San Simeon, *129*, 1936, L. S. ROSE, C.

3. Monterey Co. Jolon, *127*, 1935, D. D. KECK and P. STOCKWELL, C.

These two specimens from the southern coastal range are of a similar habit. Both are low and have somewhat arched, slender scapes. The umbels are thin, pedicels of different length. Pollen normal, *129* is diploid, *127* tetraploid, although *127* is more slender and gracile than *129*, and has fewer flowers per umbel.

4. Tulare Co. Visalia, *17*, 1898, P. S. WOOLSEY, B; 3 Rivers, *75*, 1925, L. R. ABRAMS, D; Middle Tule River, PURPUS (JEPSON, 1922).

17 and *75* deviate very much from other types of *Allium amplectens* with their long pedicels and large star-shaped flowers. They possibly belong to another species. Pollen normal, both diploid.

5. San Benito Co. Hernandez, *68*, 1903, L. M. LATHROP, D; Pinnacles, *124*, 1937, J. T. HOWELL, C.

68 is a small, delicate type with upright scapes. Pollen normal, tetraploid. *124* is tall (5 dm) and robust, thick scapes, large spherical umbels (Fig. 11 *a*). Pollen asynaptic.

6. *Mariposa Co.* Mariposa, *66*, 1889, *70*, 1892, *19*, 1893, 1894, *13*, no date, J. W. Congdon, BD; *84*, 1914, S. Fauntleroy, C; Mt. Bullion, 7 *b*, no date, S. Suney, B.

These are similar in type, erect scapes, umbels small and dense. Both normal and asynaptic pollen, types with normal pollen are tetraploid.

7. *Santa Clara Co.* Coyote Cr., *62*, *63*, 1895, W. R. Dudley, D; Stanford University, *65*, 1898, L. R. Abrams, D, *4*, *59*, *89*, 1903, A. D. E. Elmer, BCD; Uvas-Almaden Rd. *64*, 1922, R. S. Ferris, D; Gilroy Hot Spr., *111*, 1937, A. Eastwood and J. T. Howell, C; Isabel Cr., *133*, *134*, *135*, J. Clausen, D. D. Keck and A. Levan.

8. *San Mateo Co.* Jasper Ridge, *61*, 1921, H. L. Mason, D; Stanford University, *58*, 1932, L. R. Abrams, D.

The specimens from Santa Clara and San Mateo are rather consistent in type, medium-sized, normal. With the exception of some of the forms from Isabel Cr. all have normal pollen and are tetraploid (*64*, however, which deviates considerably from *amplectens*, is a diploid).

9. *Alameda Co.* Cedar Mt., *60*, *88*, *98*, 1903, A. D. E. Elmer, CD.

10. *Contra Costa Co.* Mt. Diablo, *97*, 1902, W. W. Carruth, C.

Simple erect scapes with leaves only in their lowest part. They all agree in habit. From Cedar Mt. the diploid *88* and the tetraploid *60* should be compared. The diploid is evidently larger and more robust than the tetraploid. *98* has mixed pollen, normal and asynaptic, even within one stamen, the others have normal pollen.

11. *Marin Co.* Mt. Tamalpais, *30*, 1892, T. S. Brandegee, B; Lagunitas, *15*, 1893, W. C. Blesdale, B; Kentfield, *100*, 1912, M. E. Parsons, C.


Fig. 12. The distribution of *Allium amplectens*.

30 and *100* very stout types with big bulbs, *15* more slender. All three have normal pollen, tetraploids.

12. *Sonoma Co.* Hill-sides, Sonoma, type locality of the species (TORREY, 1856); Hood's Peak, *12*, 1892, MICHENER and BIOLETTI, B, *93*, *110 b*, *130*, 1927, M. S. BAKER, C; Petrified Forest, *85*, 1915, A. EASTWOOD, C; Adobe Canon, *123*, 1927, M. S. BAKER, C; Shellville, *91*, 1931, L. S. ROSE, C.

91, *93*, *110 b* and *130* are of a very characteristic type. Groups of small rounded bulbs grow together »in bunches of 6—30», evidently in moist places. The scapes are very slender and gracile, often somewhat undulated, the inflorescences are small, few-flowered, pedicels short. These four specimens have normal pollen and are diploid. This form will be referred to later as the Sonoma-type (Fig. 11 *b*). *12* and *85* are of a more common, taller and stouter growth. Pollen normal, tetraploids.

13. *Napa Co.* Howell Mt., *18*, 1899, J. P. TRACY, B; Walter's Spr., *55*, 1933, D. D. KECK, D; Napa, *120*, *121*, 1933, J. T. HOWELL, C; Palisades, *112*, 1937, M. S. JUSSEL, C.

120 agrees completely with the Sonoma-type: »growing in dense clumps from numerous small bulbs». Similar to the earlier specimens of the Sonoma-type, it has normal pollen but is evidently a tetraploid. *121* was collected in the same region, had »stems growing singly» and was of quite another general appearance: big, marked bulbs, stout, short scapes, large flowers. It has asynaptic pollen. *18* reminds of *120* but is lower and more slender, normal pollen, diploid. *55* and *112* are both tetraploids and they are of strong and robust growth. Normal pollen.

14. *Solano Co.* Vaca Mt. (JEPSON, 1922).

15. *Lake Co.* Lakeport, *16*, 1902, J. P. TRACY, B; Lower Lake, *53*, 1902, A. M. BOWMAN, D, *86*, 1932, M. S. JUSSEL, C, *79*, *80*, 1938, A. EASTWOOD and J. T. HOWELL, C; Mt. St. Helena, *105*, *106*, 1915, A. EASTWOOD, C; Cache Cr., *54*, *103*, 1919, A. A. HELLER, CD, *56*, 1928, D. K. KILDALE, D; Kelseyville, *90*, 1924, J. B. BLANKINSHIP, C; Adams Spr., *114*, *116*, 1933, M. S. JUSSEL, C; Oak Park Spr., *6*, *57*, 1933, R. BACIGALUPI, R. S. FERRIS and I. L. WIGGINS, BD; Lake-Colusa Boundary, *78*, 1934, C. PURDY, C; Indian Valley, n. e. Lake Co. (JEPSON, 1922).

Most of these specimens represent stout, erect types. *90* and *114* are more gracile. 11 of them are tetraploids with normal pollen, 4 are asynaptic, only 1 is diploid with normal pollen.

16. *Colusa Co.* Stonyford, *74*, 1926, R. S. FERRIS, D; Williams, *117*, 1934, K. ESAU, C.

Both asynaptic.

17. *Mendocino Co.* Red Mts., *22*, 1901, A. EASTWOOD, B; *113*, 1937, A. EASTWOOD and J. T. HOWELL, C; Covelo, *52*, 1903, V. RATTAN, D, *87*, 1928, A. EASTWOOD, C; Ukiah, *108*, 1913, A. EASTWOOD, C; Hopland, 7, 1921, J. P. TRACY, B, *115*, *122*, 1936, A. EASTWOOD and J. T. HOWELL, C; Hearst, *50*, 1927, R. BACIGALUPI, D.

All agree rather well in type, *115* and *122* have unusually rich-flowering inflorescences. 6 of them have normal pollen and are tetraploids, while 3 are asynaptic.

18. *Humboldt Co.* Kneeland Prairie, *51*, 1903, V. RATTAN, D, *30 b*, 1912, *1*, 1921, J. P. TRACY, B; Bald Mt., *3*, 1923, J. P. TRACY, B; Yager, *42*, 1923, J. P. TRACY, D, *131*, 1937, A. EASTWOOD and J. T. HOWELL, C; Van Duzen River, *126*, 1936, C. C. and S. K. HARRIS, C (Plantae exsiccatae Grayanae, No. 645).

3 is a very gracile diploid with normal pollen (Fig. 11 *c*), *126*, tall, rich-flowered, asynaptic, the others are stout and tall tetraploids with normal pollen.

19. *Del Norte Co.* Gasquet, *32*, 1935, H. E. PARKS and J. P. TRACY, B.

Tall, rather slender form with normal pollen, tetraploid.

20. *Placer Co.* Auburn, *92*, 1915, E. HANVER, C; Colfax, *110*, 1932, B. R. JACKSON, C.

21. *Nevada Co.* Nevada City, *101*, 1912, A. EASTWOOD, C; Banner Hill, *23*, 1916, H. M. HALL, B; Grass Valley, *99*, 1919, A. A. HELLER, C.

All these specimens from Placer and Nevada show a great similarity in general appearance: small rounded bulbs, erect, gracile scapes, small umbels; they have all normal pollen and all, except *92*, are diploid. The tetraploid form *92*, however, does not deviate from the others morphologically.

22. *Yuba Co.* Marysville, *95*, 1930, A. EASTWOOD, C.

Differs very much from the preceding specimens: tall, stout, rich-flowering. Pollen normal, tetraploid.

23. *Butte Co.* Grain Fields, 24, 1896, R. M. AUSTIN, B; Clear Cr., *72*, 1897, H. E. BROWN, D, *69. 70 b*, 1902, A. A. HELLER and H. E. BROWN, D; Chico, 9, 1898, BRUCE, B, *8*, *73*, *102*, 1914, A. A. HELLER, BCD; Oroville, *94*, 1931, L. S. ROSE, C; Durham, *81*, 1935, F. BROWN, C; Paradise, *119*, 1936, M. E. WALL, C.

Among these an erect, rather slender type predominates. Its umbels are small, dense, capitate. *119* approaches the tufted Sonoma-type. *9* and *94* are more robust. All have normal pollen and all, except the gracile type *119*, are tetraploids.

24. *Tehama Co.* Paynes Cr., *76*, 1930, D. K. GILLESPIE, D, *49*, *118*, 1934, A. EASTWOOD and J. T. HOWELL, DC; Rosewood (JEPSON, 1922).

Normal pollen, tetraploids.

25. *Shasta Co.* Anderson, *83*, *109*, 1913, L. E. SMITH, C; Kennett, *107*, 1913, L. E. SMITH, C; Montgomery Cr., *104*, 1923, E. BETHAL, C; Redding, *82*, 1934, A. EASTWOOD and J. T. HOWELL, C.

82 and *107* tall, the others small and tender, especially 104, which is very gracile with small umbels and short pedicels. They have all normal pollen and are tetraploids.

26. *Trinity Co.* Union Cr., *25*, 1909, H. M. HALL, B; Scott Mts., *128*, 1931, A. EASTWOOD and J. T. HOWELL, C; Eagle and Bear Cr. *125*, 1937, A. EASTWOOD and J T. HOWELL, C.

25 and *128*, low, gracile, few-flowered, resemble Shasta *104*. They have asynaptic pollen. *125* decidedly more vigorous, normal pollen, diploid.

27. *Siskiyou Co.* Yreka, *28*, *71*, 1910, G. D. BUTLER, BD; Mt. Shasta, A. A. VEATCH (type locality for *Allium attenuifolium*; KELLOGG, 1861).

28 (Fig. 11 *d*) and *76*, robust, erect, both asynaptic.

28. *Lassen Co.* Susanville, *10*, 1892, T. S. BRANDEGEE, B; Dixey Mts. *11*, *31*, 1894, M. S. BAKER and F. NUTTING, B; Pine Cr., *14*, 1898, C. C. BRUCE, B.

11 and *14*, strong, robust, very large umbels, *10* and *31*, low, tender, small umbels, very short pedicels. All except *10* are asynaptic. The very striking difference in size between *11* and *31*, which were collected on the same occasion, together with the difference in pollen size makes the conclusion drawn in another connexion highly probable, that different chromosome numbers may occur among asynaptic forms as well as among types with normal pollen.

29. Modoc Co. 31 c, 1893, M. S. BAKER, B; Altura, *67*, 1919, R. S. FERRIS and R. DUTHIE, D.

67 resembles the small Lassen types, asynaptic.

OREGON.

30. Josephine Co. Waldo, 77, 1928, Kerby, *77 b*, 1928, J. W. THOMPSON, D.

77 very small and delicate, leaves filiform. Normal pollen, tetraploid.

31. Jackson Co. Rogue River, *29*, 1893, R. M. AUSTIN, B; Siskiyou Mts., *44*, 1894, F. M. ANDERSON, D; Chinquapin Mt., *47*, 1925, E. J. APPELGATE, D; Pinehurst, *38, 40*, 1927, M. E. PECK, D.

38, 40 and *47* have normal pollen, tetraploids, the others have asynaptic pollen.

32. Klamath Co. Swan Lake, *48*, 1923, E. J. APPELGATE, D; Bonanza, *39, 43*, 1927, M. E. PECK, D.

43 has normal pollen, tetraploid, the others asynaptic.

33. Lake Co. Lakeview, *46*, 1927, M. E. PECK, D.

Tall, stout, small inflorescences, asynaptic.

34. Harney Co. Steen Mt. *34*, 1896, J. B. LEIBERG, B, *33*, 1898, W. C. CUSICK, B, *45*, 1925, M. E. PECK, D.

Asynaptic.

35. Lane Co. Dorena, *2*, 1924, L. CONSTANCE, B.

Small, tender, filiform leaves, normal pollen, diploid.

36. Linn Co. Albany, *41*, 1928, J. W. THOMPSON, D.

Tall and stout, normal pollen, tetraploid.

WASHINGTON.

37. Klickitat Co. According to PIPER (1906), *Allium attenuifolium* occurs in a collection, SUKSDORF 60, from this county. In reply to an inquiry on this matter, Dr. C. W. SHARSMITH, Pullman, very kindly gave me the following information in a letter: 'After a careful search not only through the herbarium, but also through PIPER's and SUKSDORF's as yet unmounted material, I failed to find the SUKSDORF 60 specimen cited by PIPER. However, I did find among SUKSDORF's unmounted plants a single sheet containing rather scanty, albeit sufficient material for verification, of *Allium amplectens* TORR. This SUKSDORF collection was unnumbered; it is named by him *A. attenuifolia*, and is given the locality in his own handwriting as 'WS 24 May, 1881'. The 'WS' I interpret as White Salmon (in western Klickitat County, Washington), on the basis of our knowledge as to his whereabouts at this time . . . . From this it is clear that *Allium amplectens* goes as far north as to the southern parts of Washington.

3. PLANT-GEOGRAPHICAL SURVEY.

By putting together the data from the different specimens of *Allium amplectens*, listed in the preceding chapter, material is furnished of a rather detailed distribution map of the species. 120 of the specimens are from California and 16 from Oregon. To this may be added some

localities from the literature. If different specimens, which might possibly represent the same locality, are combined, the following result is reached: 68 certainly different localities in California, 10 in Oregon and 1 in Washington. These latter are the data plotted on the map (Fig. 12).

It may be seen from this map that the distribution of the species extends along the Pacific Coast from southern California into southern Washington. Its main occurrence is northern California, and its densest distribution is found in Napa and Lake Counties in California. Possibly, however, this density is only apparent, since this region may be more frequently visited by botanists. Thus Dr. H. L. MASON informs me in a letter: »Certain counties of the Sierra Nevadas have not been intensively collected, and it is extremely likely that the species occurs in all of the counties north of Tulare».

Pollen samples were examined from so many

Fig. 13. The distribution in nature of normal and asynaptic meiosis in *Allium amplectens*, rectangular dots = normal meiosis, round dots = asynaptic meiosis, rectangle within round dots = both types occur.

herbarium specimens as possible. Acetocarmine slides turned out to be very useful. It was possible in each case to decide with certainty if

TABLE 1. *The occurrence of asynapsis in the different counties.*

Normal pollen

County No.	County	Number of specimens	County No.	County	Number of specimens
2	San Luis Obispo	1	21	Nevada	3
3	Monterey	1	22	Yuba	1
4	Tulare	2	23	Butte	11
8	San Mateo	1	24	Tehama	3
10	Contra Costa	1	25	Shasta	5
11	Marin	3	30	Josephine	1
12	Sonoma	5	35	Lane	1
19	Del Norte	1	36	Linn	1
20	Placer	2			
				Total	43

Asynaptic pollen

County No.	County	Number of specimens
1	San Diego	5
16	Colusa	2
27	Siskiyou	2
29	Modoc	1
33	Lake Oreg.	1
34	Harney	3
	Total	14

Both types of pollen

County No.	County	Number of specimens Normal pollen	Number of specimens Asynaptic pollen	County No.	County	Number of specimens Normal pollen	Number of specimens Asynaptic pollen
5	San Benito	1	1	28	Lassen	1	3
6	Mariposa	2	2	31	Jackson	2	2
7	Santa Clara	9	1	32	Klamath	1	2
9	Alameda	2	1				
13	Napa	4	1				
15	Lake Calif	12	4				
17	Mendocino	6	3				
18	Humboldt	5	1				
26	Trinity	1	2				
					Total	46	23

normal or asynaptic pollen was present. Among the 126 specimens, where this examination could be carried out, no less than about $1/8$ (37 specimens) showed asynaptic pollen, while 89 specimens had normal pollen. When these conditions were plotted on the map it was found that the phenomenon of asynapsis had a very wide occurrence in nature: Among 34 counties represented, 16 had only normal pollen formation, 5 had purely asynaptic pollen and 12 had both types of
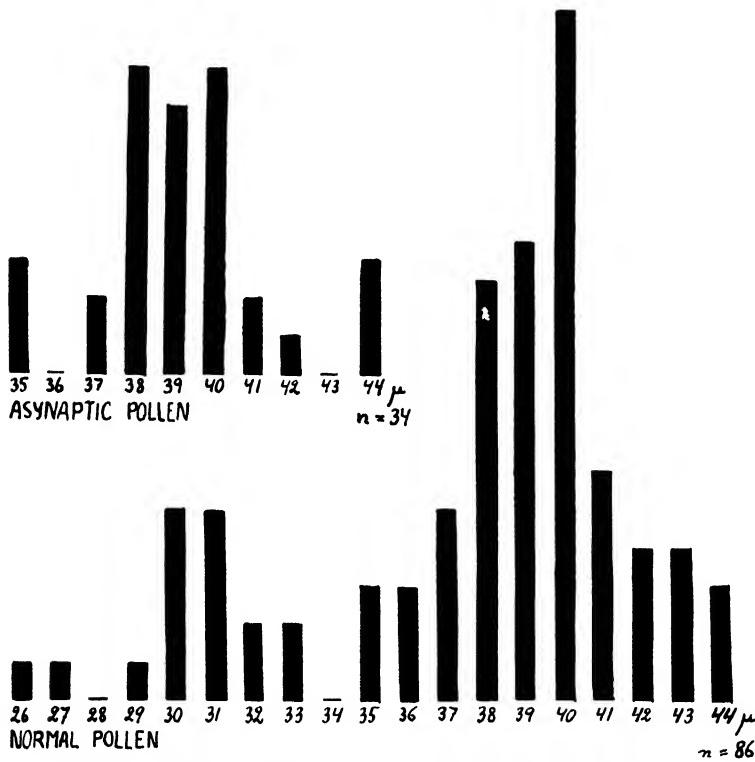

Fig. 14. Pollen length in the normal and asynaptic type.

pollen (Table 1). The map (Fig. 13) gives a summary of these conditions.

It will also be seen from the map that the asynaptic genotype is scattered over the total area of the species. Since necessarily very few specimens from each county could be examined, it is impossible to draw any far-fetching conclusions from the map. A few facts may still be pointed out although with some reservation. All specimens from San Diego are asynaptic, which makes it plausible that this locality, isolated from the rest of the distribution of the species, may contain exclusively

asynaptic types. And in the north-eastern corner of the distribution evidently asynapsis predominates (14 asynaptic specimens as against 4 normal). Normal meiosis has a cumulative district in the inner parts of California (Placer, Nevada, Yuba, Butte, Tehama and Shasta Counties) with 25 examined specimens, among which not a single asynaptic type was found. In order to get a deeper knowledge of the situation it will be necessary, however, to carry out detailed field studies.

Pollen grains were measured from each pollen sample. It is important that identical stages of development are present in the measured samples. This condition was easily fulfilled, however, since even in dried material the nuclei of the pollen are clearly visible in acetocarmine slides. I selected for measurement the stage a few days after the first pollen mitosis, when the strongest growth of the pollen grains is past. Then both the pollen nuclei could be readily seen in most slides. In some cases it was possible even to see that the specimen had been dried during the pollen anaphase, as clearly necrotic restitution nuclei had been formed in the pollen grains.


Fig. 15. The distribution of diploid and tetraploid types of *Allium amplectens*, transverse lines = diploids, longitudinal lines = tetraploids.

The averages of the pollen lengths are collected in the graph

TABLE 2. *The occurrence of diploid and tetraploid forms in the different counties.*

Diploid forms			Tetraploid forms			Both diploids and tetraploids			
County No.	County	Number of specimens	County No.	County	Number of specimens	County No.	County	Number of specimens 2x	4x
2	San Luis Obispo	1	3	Monterey	1	9	Alameda	1	1
4	Tulare	2	5	San Benito	1	12	Sonoma	2	2
10	Contra Costa	1	6	Mariposa	1	13	Napa	1	3
21	Nevada	3	7	Santa Clara	7	15	Lake Calif.	1	11
26	Trinity	1	8	San Mateo	2	18	Humboldt	1	4
35	Lane	1	11	Marin	3	20	Placer	1	1
			17	Mendocino	6	23	Butte	1	8
			19	Del Norte	1				
			22	Yuba	1				
			24	Tehama	3				
			25	Shasta	5				
			28	Lassen	1				
			30	Josephine	1				
			31	Jackson	2				
			32	Klamath	1				
			36	Linn	1				
Total		9	Total		37	Total		8	30

(Fig. 14). It will at once be seen that the normal pollen grains form a bimodal curve. This indicates that the material contains not only the tetraploid forms, which in actually cytologically examined material were shown to have a pollen length corresponding to the right mode of the graph, but also diploid forms corresponding to the left mode. The curve of the asynaptic pollen, too, shows a tendency to more than one mode. In this case, however, there is too little material to allow any conclusions being drawn.

If a pollen length of $34\,\mu$ is selected as the limit between diploids and tetraploids with normal pollen, the result in the distribution of the different forms is the one represented in the map (Fig. 15) and in Table 2. It will be seen from these data that the tetraploids are very much commoner in nature than the diploids: among 84 specimens examined, 67 were tetraploid and 17 diploid. 6 counties had only diploid forms, 16 had tetraploid forms and 7 had both types.

4. THE CORRELATION BETWEEN POLLEN TYPE AND MORPHOLOGY.

Two morphological properties, plant height and pollen fertility, were plotted against pollen type. Table 3 gives the results concerning plant height. It will be seen from the table that the plant height is decidedly lower in the diploids than in the tetraploids and the asynaptic forms. On the other hand, there is no significant difference between the asynaptic plants and the normal tetraploids. In various places in the list of the herbarium specimens I pointed out that a difference in height and in general viability running in the wrong direction could be found if certain individual diploid and tetraploid specimens were compared. It is clear that the genic variation in these cases goes in the opposite

TABLE 3. *Pollen type and plant height.*

Pollen type	Plant height in cm.																		Total	Medium
	19	21	23	25	27	29	31	33	35	37	39	41	43	45	47	49	51	53		
Normal {2x	1	1	1	6	1	—	1	5	—	—	—	—	—	—	—	—	—	—	16	27,8
Normal {4x	3	2	2	7	7	7	9	5	7	7	7	2	2	2	—	—	—	3	63	33,2
Asynaptic	1	4	4	3	2	3	2	4	3	3	1	3	1	—	1	—	—	2	34	31,5

direction to the genomatic variation. This source of error is diminished
if a sufficient number of individuals are collected within each group.
The genic variation within the groups cannot under such circumstances
conceal the difference between the groups.

In Table 4 the pollen fertility of the different pollen types is
examined. In spite of the fact that some diploids had very bad pollen
(in reality triploids?), the diploids turned out to have, on the average,

TABLE 4. *Pollen type and pollen fertility.*

Pollen type	Percentage good pollen											Total	Medium
	0	10	20	30	40	50	60	70	80	90	100		
Normal {2x	—	—	1	1	1	—	1	2	1		9	16	79,₄
{4x	1	1	2	4	9	12	7	17	7		4	64	61,₇
Asynaptic	1	3	2	2	—	3	3	8	8		2	32	62,₇

better pollen than the other groups. More than half of the diploid
plants had more than 90 % good pollen, a fertility rarely occurring
among the other types.

It must be considered of interest that a difference between the
normal diploids and tetraploids could be demonstrated even by these
rather crude methods. It affords a certain support to the method of
classification of the pollen; it shows, in my opinion, that the division
into diploids and tetraploids, made according to the pollen size, really
represents valid conditions.

III. DISCUSSION.

The term *asynapsis* usually denotes a whole group of phenomena,
characterized by a decreased chromosome pairing during the first
meiotic division. Asynapsis can be modificative or genotypic. Tem-
perature, age, chemical action may cause asynapsis. Evidently failure
of chiasma formation is the typical response, if a certain stage of meiosis
is subjected to external irritations. The genotypic asynapsis, on the
other hand, may be due to lacking chromosome homology, that is the
case in numerical and structural hybrids and haploids, and it may be
due to a true gene action.

In all probability the case of *Allium amplectens* belongs to this
last category. By studying the earlier, best known cases of such asyn-
apsis, viz. *Zea* (BEADLE, 1930, 1933), *Datura* (BERGNER, CARTLEDGE and
BLAKESLEE, 1934), *Crepis* (RICHARDSON, 1935), *Pisum* (KOLLER, 1938),

and *Nicotiana* (GOODSPEED and AVERY, 1939), it should be possible to obtain a picture of the normal course, along which the genically caused asynapsis takes place. I shall now examine to what extent the *Allium* case agrees with this normal course and to what extent deviations occur.

The chromosome pairing at zygotene is normal in the genically caused asynapsis cases. This has been actually observed in *Zea, Crepis* and *Nicotiana.* And this condition is even more evident in *Allium*, where the prophases are especially clear. It has been demonstrated beyond doubt that the chromosome pairing at pachytene is morphologically as intimate and complete in the asynaptic as in the synaptic types. It should be mentioned, however, that in an asynaptic *Rumex acetosa,* studied by YAMAMOTO (1934), a clear difference in zygotene pairing was observed as compared with synaptic types.

It is evident that no conclusions can be drawn concerning the formation of chiasmata from the appearance of the pachytene, if the pairing is good. This is seen in those *Allium* forms which have their chiasmata localized to the centromeric region. They have apparently quite regular pachytene pairing along the whole length of the chromosomes. And in a tetraploid species with such chiasma localisation the frequency of quadrivalents is great at pachytene and early diplotene, while it is almost nil at metaphase I. This is explained by the condition that only the pairing close to the centromeres gives rise to chiasmata (LEVAN, in the press).

According to DARLINGTON (1937 and elsewhere), the pairing which predisposes to asynapsis, involves a change in the precocity conditions of the meiotic prophase, i. e. a premature division of the chromosomes, which impedes true pairing and leads to the formation of chiasmata only in exceptional cases. The pairing that remains in such chromosome regions is therefore not due to chiasmata but to the torsion of the chromosomes.

In most earlier cases of asynapsis the suppression of chiasmata is never total, but a certain, by no means small percentage of chiasmata originates. This percentage is liable to great variation in the same individual, all values from 0 to 100 % may occur. In *Zea* an average of 3—82 % bivalents is formed in 8 plants examined, in *Crepis* about 50—70 % of the potential bivalents were realized, and in *Pisum* the corresponding values were of a similar magnitude (at diakinesis 71 %, at metaphase 54—64 %). In this respect *Allium* is very strikingly different from earlier cases, the asynapsis of *Allium* being very much more complete. One chiasma in some 500 cells is typical and the

highest frequency of chiasmata observed is 4 chiasmata in 100 cells. This corresponds to a percentage of realized chiasmata counted on the potential chiasmata of only about some 100th per cent.

The only previously described asynapsis case which in this respect is comparable to *Allium* is *Datura*, where often 24 univalents were found at metaphase I. Their behaviour was quite irregular, however, and they were not collected in any equatorial plate.

The lack of chiasmata in *Allium* at metaphase I must be considered rather remarkable, if viewed in connexion with the complete pachytene pairing. There has often been a tendency in the literature to interpret the few chiasmata which are formed in the asynaptic species as corresponding to the amount of pairing seen at pachytene. Thus DARLINGTON (1937, p. 405) writes: »Where failure of chiasmata is absolute, pachytene is also very defective. Where it appears complete, chiasmata are always formed in a proportion of nuclei, a proportion subject to great local variation». The conditions in *Allium* show, however, that it is not necessary for an apparently normal pachytene pairing to lead to the formation of any chiasmata.

A common feature in all earlier cases of asynapsis is that the spindle never acquires any command over the univalents. While the few bivalents may often be arranged into an equatorial plate, the univalents remain irregularly scattered out over the whole length of the spindle. The spindle itself behaves characteristically, it grows in length and is often bent round in the cell so that its poles approach each other (cf. the behaviour of the spindle in asynaptic *Drosophila pseudo-obscura*). In *Allium* there is no trace of all this. The first division is above all characterized by the complete control of the spindle over the situation. The spindle is regular, bipolar. The univalents and the solitary bivalents are moved towards the equator and arranged into one plate or, in addition, into one or two accessory plates. The bivalents have the same type of orientation as the univalents.

The cause of this striking difference from other cases of asynapsis must be found in the behaviour of the centromeres. They must in *Allium* have the same structure both in univalents and in bivalents. They must all be polarized and capable of auto-orientation. This is no doubt the cause of the great regularity with which the first division takes place, and this also causes the difference from earlier cases of asynapsis and also the behaviour of univalents in general, which owing to lack of partners and lack of centromeric polarisation are devoid of the possibility of auto- as well as co-orientation. Ordinary univalents,

it is true, very often later on, after the separation of the bivalents, proceed towards the equator and orientate themselves into a secondary equatorial plate. This may be expressed in this way: their centromeres were unpolarized from the beginning, but later on became polarized. In *Allium*, on the other hand, the polarisation must occur very early, at the latest immediately after the disappearance of the nuclear membrane.

The cause why the centromeres in *Allium*, in spite of their polarisation, remain undivided through the first division is unexplained. It must be seen in connexion with the rhythm of meiosis, where the centromeres normally have to play a part in two nuclear divisions without being divided more than once. In the c-meiosis (LEVAN, 1939) the contrary behaviour is present, the chromosomes separate in the first division and soon after this the centromeres divide without any interkinesis despiralisation. I attributed this behaviour to the lack of pressure on the centromeres of the interior spindle, which should involve a preparation for a rapid division of the centromeres. The conditions of *Allium amplectens* argue against this assumption. No pressure of any interior spindle is present and the centromeres are still undivided.

Abruptly, while the chromosomes are being prepared for the anaphase I, the telophase starts, and the chromosomes still on the equatorial plate are included into a disciform interkinesis nucleus. No directly comparable case to this behaviour seems to be known. ROSENBERG's (1927) semi-heterotypic division has the same end-result but its course is very different.

Immediately after the interkinesis follows the second division, during which the chromosomes are less contracted than during the first division. Worth mentioning is the observation that no accessory plates, which are almost regularly present at the first division, were seen during the second division, all the centromeres being very regularly arranged in the equatorial plane. Since the chromosomes are really much larger during the second division than during the first it seems improbable that mere crowding of the plate should be the cause of the failing congression of the chromosomes in the first division. Besides that, non-congression has hardly been observed earlier in chromosomes with polarized centromeres. Instead it is found among bivalents, which, due to crowding and body-repulsion, are prevented from arranging themselves in the first metaphase plate. The situation in *Allium amplectens* goes to show that it cannot be just the conditions of the centro-

meres, but rather the special conditions of the first metaphase, which cause the formation of accessory metaphase plates.

The result of meiosis is dyad pollen with somatic chromosome sets. Apart from the first division, which did not lead to any chromosome separation, a certain agreement is noted with the deviation of meiosis, described by GUSTAFSSON (1935) in apomictic *Taraxacum* and called pseudo-homeotypic division. Its end-result is also unreduced dyads. The behaviour of the univalents, which are gathered in an equatorial plate, is ascribed to their bipolarity: »Owing to the failure of the spindle to stretch and the failure of the nuclear membrane to form, the spindle attachments of the univalents are able to divide and the chromosomes, as a result of their bipolarity, actively move towards the equatorial plane, where the halves separate» (l. c. p. 81).

From the above discussion it will be seen that the asynapsis of *Allium amplectens* is of a greatly different course from the cases of asynapsis described earlier, although it clearly belongs to the same type as *Zea*, *Datura*, *Crepis*, *Pisum* and *Nicotiana*. The regularity of the whole process makes the *Allium* asynapsis comparable to other gene-controlled deviations of meiosis. The closest resemblance exists, as mentioned above, to the monokinetic meiosis (LEVAN, 1935), where the first division occurs normally but the second is omitted. In this abnormality the mechanism is quite different, however, chiasmata being formed in a normal manner and the pre-reduced parts of the bivalents separating at the first division. The second division is lacking, no chromosome contraction and no spindle formation are seen. It is substituted by a long resting stage. The result is, generally speaking, the same as after asynapsis: pollen dyads with doubled chromosome number. Since the first division takes place, meiotic abnormalities due to autopolyploidy occur (and most monokinetic types are autopolyploid). The result of this is that the chromosome complement of the pollen varies, and in no way is so well-balanced as the asynaptic pollen. The monokinetic pollen is also distinguished from the asynaptic pollen by purely morphological characters, its chromosomes being arranged in pairs. The frequency of monokinetic pollen is very seldom exactly 100 %, often a few normal pollen grains occurred even in the most completely monokinetic types. Asynapsis, on the other hand, is almost always absolute. Only in a couple of herbarium specimens was the simultaneous occurrence of normal and asynaptic pollen observed in the same plant. And in these cases it is quite possible that the dyads were essentially of monokinetic origin.

Unfortunately I have not yet been able to study these gametic anomalies on the female side. And in *Allium amplectens* I have, owing to difficulties of cultivation, not been able to grow any progenies. The vegetative propagation is, however, so lively in *Allium* that even types with unbalanced meiosis and subsequent bad seed fertility may acquire a wide distribution in nature. The pollen size of *Allium amplectens* shows that the tetraploid is probably the maximal chromosome number occurring in nature. This indicates that if the asynaptic type is seed fertile, some regulative mechanism must be present which keeps the chromosome number down, for instance, apogamous development of the embryos.

At any rate it was demonstrated that asynaptic meiosis is widely distributed within the area of the species. The species is very multiform and evidently consists of a multitude of ecotypes more or less specialized for certain exterior conditions (for instance, the San Diego form, the Sonoma form, the Nevada form). It would be interesting to find out if the asynapsis gene in its infiltration of the species has any preference for certain morphological types. As far as can be known at present, this does not seem to be the case. Plant height, for instance, a property which is fairly characteristic of different ecotypes, exhibited a similar variation in the asynaptic types to that in the normal. Nor does the asynapsis gene seem to have any preference for certain ecological or geographical habitats. A closer study of these problems would probably give interesting results.

The fact that the asynapsis gene does not seem to have any selective value, either positive or negative, points to a vegetative or apomictic type of propagation, in which case a meiotic quality is of no importance (cf., for instance, the great distribution in nature of seed sterile triploid *Allium carinatum*).

Concerning the question of polyploidy in *Allium amplectens* the conditions are somewhat different. There is evidently present a clear difference in plant height and probably also in general viability between diploid types and tetraploids. The tetraploids are superior in the selection, they are of a commoner occurrence in nature. Conditions are often similar in other *Allium* species. The tetraploid *Allium oleraceum* is more viable than the related diploid *Allium paniculatum*. In *Allium Schoenoprasum* the diploids predominate in nature, but the only known autotetraploid is a giant form. In *Allium nutans* the tetraploids are, on an average, more robust and viable than the diploids.

It is possible that the tetraploids of *Allium amplectens* represent

the optimal stage of genomic viability of the species. It is also possible, however, that the triploid is an equally vital type to the tetraploid or even somewhat superior. Among the asynaptic types the triploids seem to be most common. All the asynaptic forms, in which the chromosome number was directly determined, were triploids, and in the graph of the pollen size (Fig. 14) the middle mode is the largest one. This might be due to an apogamous seed formation, which in the asynaptic types preserves the triploids, while among types with normal meiosis the triploids, owing to sterility, would be doomed to failure.

SUMMARY.

Allium amplectens TORR. is an endemic of the North-American Pacific Coast region. Its cytological conditions are of specially great interest since a genically controlled asynapsis is of common occurrence among its natural populations.

The first part of this paper deals with the cytology of the normal and asynaptic forms of the species. Its basic chromosome number is 7 and the chromosome morphology agrees with previously studied *Allium* species of the 7-series, with the important exception that no satellited chromosome is present. This is connected with the peculiar conditions of the nucleoli, which are formed at several chromosomes of each genome. Meiosis of normal and asynaptic forms is described. The asynaptic forms show the following interesting features: The zygotene pairing is normal, the failure of chiasma formation is nevertheless almost complete, 1 chiasma in 500 cells being formed. At the first metaphase the spindle functions normally and all the univalents are arranged at the equator. The centromeres are not divided and the metaphase goes directly over into a uninuclear interkinesis. At the second division the centromeres divide, which is followed by a normal anaphase, giving rise to pollen dyads with somatic chromosome complements. Besides the dyads there are also found single monad pollen grains, where both meiotic divisions have been omitted, due to the added action of asynapsis and monokinetic meiosis.

The second part is based on studies of herbarium material. The distribution in nature of the species is determined. Further, pollen samples from the herbarium specimens are examined and the occurrence of asynapsis as well as of diploid and tetraploid forms is recorded on the map. Finally, the asynapsis of *Allium amplectens* is discussed in relation to earlier cases of asynapsis.

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CONTENTS.

A HYBRID BETWEEN TRITICUM TURGIDUM AND AGROPYRON JUNCEUM

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BOTH Russian and Canadian workers have successfully crossed wheat with *Agropyron junceum* (TSITSIN, cited by VERUSHKINE, 1935, p. 8; JOHNSON, 1938, p. 240).

The female parent of the hybrid studied by me was *Triticum turgidum* L., »Rivet's Bearded». Pollen was obtained from a wild population of *Agropyron junceum* (L.) P. B. growing at Råå in South Sweden. 245 cross-pollinated flowers gave 38 seeds (15,5 %). The seeds were germinated in petri-dishes containing sand and water. Three plants were obtained. Some more seeds germinated, but the resulting seedlings died at an early stage.

Only one of the plants flowered in 1939 (it had been germinated in the autumn of 1938). Two plants germinated in the spring of 1939 have not flowered yet.

The morphology of *Triticum turgidum* × *Agropyron junceum* seems to be rather interesting, this hybrid being more similar to wheat than most *Triticum-Agropyron* hybrids described in literature. This is especially true of the spikes (Fig. 1). These statements are based on the general appearance of the hybrid. The inheritance of the individual characters has as yet been observed only in a few cases. The

Configuration		Number of cells	
Univalents	Bivalents	Fixation a	Fixation b
14	7	1	2
16	6	4	7
18	5	8	11
20	4	8	3
22	3	2.	2
24	2	1	—
28	0	1	—
Average a	19,12	4,41	Total 25	25
» b	17,68	5,16		
» of a and b	18,4	4,8		

beards of the wheat parent are recessive (although not completely). The pubescence of the spikelets is dominant. *Agropyron junceum* has its nodes concealed within the vaginae, and so has the hybrid. The leaves of *A. junceum* have on their underside a sub-epidermal layer of mechanical tissue. This character is recessive. It is not known as yet

Fig 1   Spikes of the material   From left to right: *Triticum turgidum*, the F_1 hybrid; *Agropyron junceum*

if the hybrid is perennial.   Its morphology will be treated more in detail in a later paper (e. g. in conjunction with the descendants of the hybrid if such can be obtained).

The root tips and the anthers were fixed in chrome-acetic-formalin. The anthers were prefixed in 96 % alcohol.   The preparations were stained with gentian violet.   The three hybrid plants had 2n = 28, like both parents.   Two fixations of the plant which flowered gave the

frequencies of univalents and bivalents at first metaphase tabulated above (p. 395).

First metaphase was observed also in an E.M.C. The configuration could not be analysed, but seemed to be similar to those observed in the P.M.C:s.

Most chiasmata are terminal, but some are subterminal. Most bivalents are rod-shaped. In the cell presumably having 0_{II} the configuration is not quite clear. In a few cells some possible quadrivalents were observed, but these might perhaps be composed of two bivalents.

Assuming that the chromosome pairing is not reduced by other causes than an incomplete homology between the chromosomes, the conclusion can be drawn that *A. junceum* is not completely autopolyploid. This observation does not prove with certainty, however, that the quadrivalents observed by me in *A. junceum* (ÖSTERGREN, 1940) are due to segmental interchange.

Autosyndesis between the A- and B-genomes of wheat can probably

Fig. 2. Microphotograph of a cell with 7_{II}. — $\times 1400$.

account only for a minor part of the pairing found in this hybrid. The chromosome pairing in haploid *T. durum* was studied by KIHARA (1936). The chief causes of the pairing in my hybrid should then be autosyndesis between the *junceum*-chromosomes and allosyndesis. Allosyndesis or autosyndesis may prevail, or both may occur to about the same extent.

In two pollen tests, containing more than 1000 grains, not a single good grain was found.

The relatively pronounced similarity of this hybrid to wheat may be of interest from a practical point of view. Its high sterility does not make it very probable, however, that an F_2 can be obtained. Perhaps it may give rise to an amphidiploid.

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STUDIES ON THE SIGNIFICANCE OF POLYPLOIDY

IV. OXYCOCCUS

BY *O. HAGERUP*

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A NYONE who has had the opportunity of studying the growths of *Oxycoccus*, which adorn the *Sphagnum* bogs of our country, will have noted that this small shrub varies greatly; thus, for instance, its fruits are of different shape, size, and colour. Very conspicuous, also, is the variation in thickness (Figs. 2—3), shape, and size of the leaves (Figs. 7—9), and the manner in which the margin is recurved. Often the leaves are almost vertically erect, but they may also be horizontally expanded (Fig. 1). The flowers, too, differ in a number of characters, of which their number and the pubescence of the stalk, in particular, are of taxonomic value.

If, in addition, one has the opportunity of examining abundant material derived from ecologically different localities and from other countries, the various forms will be found to differ also in ecological and geographical respects.

These characteristic differences in morphological, ecological, geographical, and systematical characters are combined in an interesting way with equally characteristic cytological features, which will be described in detail below, as far as the available material permits.

My material consisted of a fairly large collection of dried plants belonging to the Botanical Museum of the University of Copenhagen and collected in different countries; but in addition I had collected an abundant living material in several bogs in the vicinity of Copenhagen and in Greenland near 66° N. lat.

It is of great value that several investigators have collected cytological material for me of foreign types, difficult to obtain. For this reason I owe special thanks to Dr. A. E. PORSILD, Canada, and to Mr. G. THORLÁKSSON, B. Sc., who fixed *O. microcarpus* in North Iceland.

The chromosomes are excellently stained by the method of FEULGEN.

The taxonomy has been well treated by SAMUELSSON, M. P.
PORSILD, and A E. PORSILD, whose views and nomenclature I adopt
in the following account   Most of the floristic manuals, however, are
almost useless in an attempt to ascertain the geographical conditions,

Fig 1   Top *O microcarpus* from N Iceland (66° N lat)   Middle *O quadripetalus*
var *microphyllus* from West Greenland (66° N lat)   Base *O gigas* from the
Lyngby bog near Copenhagen — Natural size

partly because of the confusion of the nomenclature, and partly be-
cause the authors have not been sufficiently interested in the varieties
On that account it is possible at present to give only the main features
of· the distribution of the various forms; thus the geographical con-
ditions are badly in need of a further elucidation, so that the dis-
tribution of the different forms may be mapped

Oxycoccus microcarpus TURCZ. (n = 12). — In nearly all its parts the plant proves to be the smallest representative of the genus. That it should be considered an independent species has already been shown by SAMUELSSON, who had a large material at his disposal, both from Scandinavia and the Central European Alps, where the plant is widely distributed and is distinctly separated from the other *Oxycoccus* forms, with which it rarely forms hybrids.

Apart from its small size, *O. microcarpus* is characterized by the shape of its leaves, but especially by being the only species with a glabrous flower stalk. In addition, the plant is recognisable by means of several other characters, which have been described in detail by SAMUELSSON. Moreover, its chromosome number is n = 12 (Fig. 7), a number not found in any of our other *Oxycoccus* forms, whereas this is the chromosome number most commonly found within the *Bicornes*.

Finally, it may be mentioned that it flowers one or two weeks earlier than *O. quadripetalus*.

Of special interest, however, are MELIN's investigations of the ecological conditions of the plant. MELIN found that *O. microcarpus* is very exclusive in the choice of its habitat, which differs from those of the other forms, in that it is only found in the driest *Sphagnum* bogs characterised by the occurrence of *Sph. fuscum, Hypnaceae*, and other xerophilous species.

The geographical distribution also is of interest. Thus, with regard to its distribution in North America, A. E. PORSILD writes as follows (1938, p. 117): »In sub-arctic *Sphagnum*-bogs only. From Alaska to the east shore of Hudson Bay penetrating but a short distance north to the limit of trees. From the Yukon Territory south through the mountains of British Columbia and Alberta. Throughout northern Alaska and the Northwest Territories *O. microcarpus* is the only representative of the genus».

The same thing may be said of the occurrence of the plant in Europe and Asia. It is circumpolar and is the species found farthest north.

In Iceland the only *Oxycoccus* species that has been found is *O. microcarpus*, which is known from a few scattered localities, especially along the north coast of the island. It is therefore all the more curious that the species has not been recorded from the nearest part of the east coast of Greenland or from the Faroes.

The species is widely distributed in northernmost Scandinavia. It decreases rapidly southward, but is recorded with certainty from the

areas round the Baltic as far as the North German moorlands (SAMUELSSON, 1922, p. 258). It has not yet been found in Denmark, but should be more carefully searched for, since it is found in the neighbouring countries both to the north and to the south.

SAMUELSSON (pp. 257—58) has also found the plant in several places in the mountains of Central Europe; thus *O. microcarpus* has a similar geographical distribution to that of the bisexual *Empetrum hermaphroditum.*

The southern limit of the distribution of the species is almost
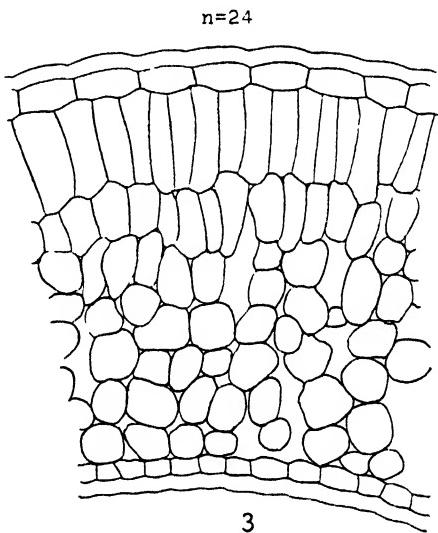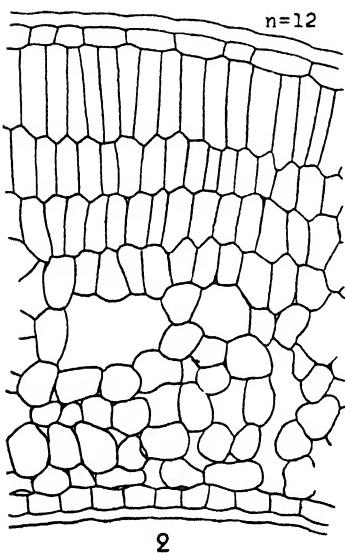
Fig. 2.   Cross section of leaf of *O. microphyllus* from N. Iceland. — × 220 —
Fig. 3.   Cross section of leaf of *O. quadripetalus* from Denmark. — × 220.

entirely unknown.   However, in the Botanical Museum of this city there is a specimen of *O. microcarpus* (from »Flora Romaniae exsiccata, No. 1085 b») collected in Distr. Ciuc in Transylvania, 1050 m. above sea-level.   This finding so far south elicits a desire to investigate the distribution of the species in all European countries.   For this purpose the floristic manuals should start from SAMUELSSON's and PORSILD's conception of the species; thus our desire is justified both by cytological, ecological, and geographical conditions.

Oxycoccus quadripetalus GILIB. var. *microphyllus* (LANGE) M. P. PORSILD.   (n = 24). — Very similar in habit to *O. microcarpus* (Fig. 1), for which reason the two forms have mostly been confused, so that it

is now impossible to unravel their distribution by means of the literature.

It was only by M. P. PORSILD's investigations (1930) that the taxonomy of this variety was elucidated so that it may now be distinguished from the other small *Oxycoccus* forms.

That each of these forms belongs to its particular species is evident,
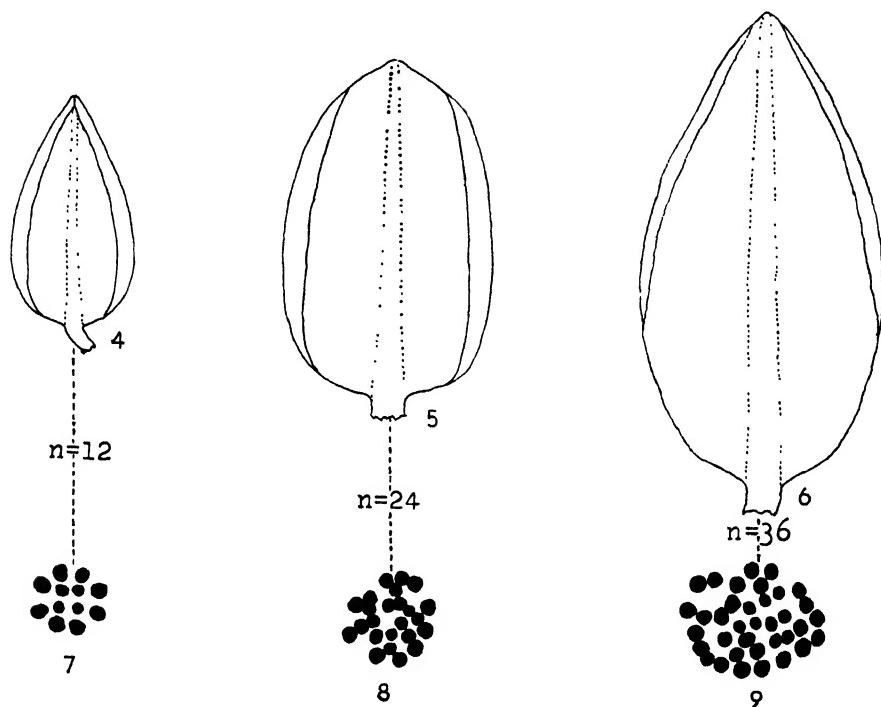
Figs. 4—6. Leaves (seen from the under side) of *O. microcarpus* (Fig. 4) from Iceland; *O. quadripetalus* var. *microphyllus* (Fig. 5) from W. Greenland (66° N. lat.); *O. gigas* (Fig. 6) from Denmark. — ×8. — Below the leaves the corresponding chromosomes in Metaphase I. Fig. 7 from Iceland, Figs. 8—9 from Denmark. — ×2500.

among other things, from the fact that the flower stalk is densely pubescent in var. *microphyllus*, which is further characterised by a different leaf shape (Figs. 4—5) and by various other characters, recorded by PORSILD. Finally, each of the two small forms has its particular chromosome number, $n = 24$ in var. *microphyllus* (Fig. 8), which I found in individuals from three different localities in nothern Seeland. However, var. *microphyllus* from Greenland is not quite identical with the Danish one, and it should be ascertained whether

they have the same chromosome number. The Greenland plant has a short style and pollinates itself (M. P. Porsild), the species thus having a tendency to develop pure lines in northern localities.

The plant is not found either in the Faroes, Iceland, or East Greenland, while it is recorded from some scattered localities along the west coast of Greenland from the southernmost point (c. 59° N. lat.) right up to a little north of the Polar circle (68° 41′ N. lat.). From this coast its range extends across to North America, of which A. E. Porsild says as follows (1938, p. 117): »On the Labrador coast from Strait of Belle Isle north to 56° 16′ N. Also in Newfoundland and the Gulf of St. Lawrence region. South of Newfoundland and the Labrador coast gradually merging into the preceding» (= O. quadripetalus).

In Denmark the species has been found in many places; it is almost as common as the main species. Often the two forms grow together, and if so, they are evenly intermixed or remain fairly isolated in small growths. However, pure growths of one or the other species may also be found, for instance, in the Bøllemosen near Copenhagen, where a large isolated pure culture of var. microphyllus is found.

That the Danish forms differ somewhat ecologically may be seen when they grow near each other but in isolated patches, as is the case in the Lyngby bog near Copenhagen. For, in such instances, var. microphyllus behaves in the same exclusive manner as O. microcarpus and chooses the highest and driest Sphagnum tufts, being more xerophilous than O. quadripetalus. It may even rise to the level of Calluna and Empetrum, but does not tolerate too much shade from other plants. On the other hand, it does not thrive in the immediate neighbourhood of water (as the main species does).

In its natural habitats the plant is recognisable in that it covers but a small part of the substratum, which is due in the first place to the very small size of the leaves, but especially to the almost erect position of the greater number of the leaves as in true light plants. And in the flowering period the plant is especially conspicuous because the very small flowers are mostly single and of a very dark colour.

Var. microphyllus differs from O. quadripetalus in all these characters, and even more distinctive properties might be mentioned (such as the size of the fruit, coloration, etc.); on seeing the two forms together, one's first impression, therefore, is that they are two different species.

There are, however, a few weighty reasons against this very

natural view; firstly, a typical *microphyllus* shoot may sometimes be found to have developed as a lateral shoot from another shoot distinctly belonging to *O. quadripetalus.*

Moreover, it is of decisive importance that these two forms have the same chromosome number, viz. n = 24. I therefore prefer to adopt the view held by M. P. PORSILD that *microphyllus* is only a variety.

As regards the geographical distribution, very little is known in addition to what has been stated above; and unfortunately only scanty and unreliable information can be derived from the literature. Although, perhaps, we are only concerned with a variety, its ecological as well as its geographical conditions seem to be well worth a closer investigation. On the whole, however, var. *microphyllus* appears to be a more northerly type than the main species. Its southern limit its entirely unknown.

Oxycoccus quadripetalus GILIB. (n = 24). — This name denotes the »main species», which is so well known that there is no need of describing it in detail here. Compared with the two preceding species it is typically much larger — even though transitional forms to var. *microphyllus* are of common occurrence. The leaves are, as a rule, large; many of them are horizontally expanded, and accordingly it tolerates a higher degree of shade than the small-leafed arctic forms, which require much light.

Its need of a certain degree of moisture is not so marked either as, for instance, that of *O. microcarpus*; it can therefore thrive right out to the open water, and does not perish until it is submerged. It extends towards dry land until it is shaded by, for instance, *Calluna* and *Empetrum*; it will not thrive, or will soon die, in the shadow of trees (e. g. *Betula*).

It would be of interest to establish its northern limit and to compare it with corresponding conditions of the small-leafed forms. It is not found either on the Faroes, in Iceland, or in East and West Greenland, but it is of common occurrence in North America, Europe, and Asia, where it decreases considerably in frequency towards the south. Its distribution in Denmark has been mapped by BÖCHER.

O. quadripetalus is tetraploid (n = 24), and in this connection it should be mentioned that both geographically and ecologically the plant has thus a higher amplitude than *O. microcarpus* (n = 12).

Finally, it should be mentioned that between the three *Oxycoccus* forms referred to above, there is no difference in the size of stomata or pollen grains. Nor did I observe any differences in the anatomy

of the wood, whereas the leaves are often relatively thicker in *O. micro-carpus* (see Figs. 2—3).

Oxycoccus gigas (=*O. microcarpus* × *O. quadripetalus?*). (n=36). — At first sight this form bears so great a resemblance to the ordinary *O. quadripetalus* that it may be difficult to distinguish them from each other without the use of a microscope. But if one has a sufficiently large material at one's disposal, *O. gigas* as a rule proves to be larger than the three *Oxycoccus* forms mentioned above; and many specimens grow to a size that *O. quadripetalus* will never attain; an individual from Finland even bore leaves measuring 0,7 × 1,5 cm.

Fig. 1 shows a specimen of the average size, and Fig. 6 a leaf; the shape of the leaf often differs somewhat from that of the other species.

The flowers are large, several together, and have long, hairy stalks. Fig. 1 further shows a characteristic feature, viz. that *O. gigas* develops vertical shoots whose leaves are more or less distinctly horizontally expanded.

My living material was collected in the Lyngby bog near Copenhagen, where the plant grows in abundance and shows a remarkable power of forming continuous carpets, which on account of the erect shoots and its entire mode of growth reminds one to a certain extent of *Vaccinium vitis idaea*. Often the carpets are so dense that only little or no space is left for other phanerogams, and even the mosses may be ousted by its shadow. The other *Oxycoccus* species, however, do not as a rule entirely cover the moss tufts, across which they spread passively, and only a minority of the shoots grow upwards. The stems are comparatively short, and the whole plant so robust that it acquires a different appearance.

Ecologically, too, *O. gigas* is quite unique, of all the forms it is the one that can endure the highest degree of drought; and it is often met with in the shape of dense, erect shrubs on the top of the moss tufts of the bog. It is also fairly well equipped against shade and can thrive below solitary trees (e. g. *Betula*); in such cases its leaves are horizontally expanded and grow big and broad. All the other forms tolerate only very little shade.

In Denmark the plant has been found as a rarity both in Jutland and on Seeland. Further, specimens from Finland and East Prussia are found in the Botanical Museum of Copenhagen. Otherwise the geographical distribution of the plant is unknown, but it would be of interest to have the matter cleared up.

Judging from the morphological and ecological conditions, it

would be natural to assume that *O. gigas* is a polyploid. And that this is actually the case will be shown more in detail below.

In the Lyngby bog near Copenhagen meiosis takes place at the end of May and the beginning of June, when the winter bud begins to grow and open, but before the flower bud has left it.

During meiosis only the first stages are fairly regular; in metaphase I (Fig. 9), however, the chromosomes are situated in such a way that it is possible to count them and to ascertain that the number is 36. Accordingly, *O. gigas* is hexaploid.
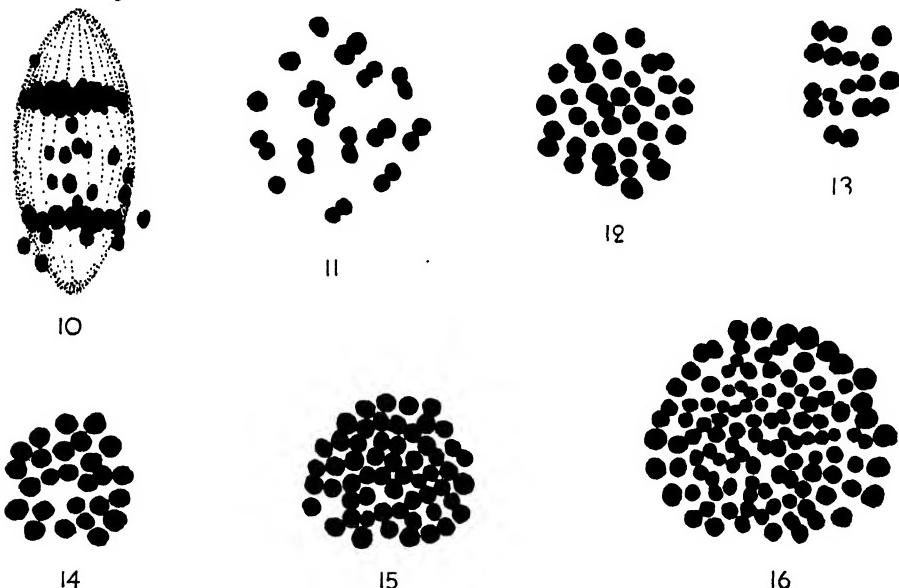

Figs. 10—16. *O. gigas*. Fig. 10, Anaphase I. Figs. 11—16, Metaphases from the first division in different microspores. In Fig. 11, n = 28; Fig. 12, n = 36; Fig. 13, n = 18; Fig. 14, n = 26; Fig. 15, n = 56; Fig. 16, n = 112. — ✕ 2880.

After metaphase I the division stages become more and more irregular, as if the plant were a hybrid. Thus Fig. 10 shows an anaphase, in which it will be seen that the chromosomes do not keep company; and a greater number of them do not enter the two daughter nuclei, but place themselves entirely outside; in the later stages the chromosomes gather in larger or smaller groups and give rise to the formation of dwarf pollen, the size of which will depend on the number of chromosomes contained in its nucleus.

In still later stages, meiosis is so irregular that the number of the chromosomes cannot be ascertained, and only by means of FEULGEN's

reaction is it possible to follow the further development of the process. In consequence of these irregularities, it frequently happens that each PMC does not form four pollen grains, as is the case in all the other species. And if we examine the contents of a young anther, we shall find, in addition to some apparently normal tetrads, many other »tetrads» which may contain 1—10 pollen grains (Figs. 17—25).

The chromosome sets of such irregular pollen grains (Figs. 17—25) may be examined by means of a careful staining with hæmatoxylin. The metaphase must then be looked for during the first division in the microspore. This division takes place when the flower bud is
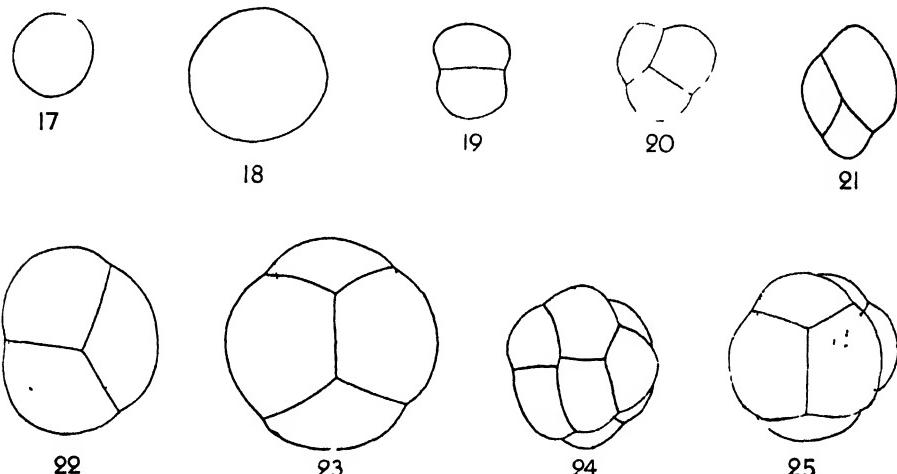
Figs. 17—25. *O. gigas.* Abnormal »tetrads» with different numbers of pollen grains. — × 850.

half-developed; it is then 2—3 mm. long, which stage may be found at the beginning of June.

As might be expected, the pollen grains (Figs. 11—16) prove to contain a greatly differing number of chromosomes. In some of the figures it is distinctly seen that, as is often the case in polyploid cells, the chromosomes have a tendency to gather in groups of two, three, or more.

Fig. 13 shows that a small pollen grain may contain 18 chromosomes, and Fig. 16 shows 112 chromosomes from a large pollen grain; it may also be possible to find all the figures from 1 to 4 × 36 in the metaphases of the pollen grains, which are mostly regular and fairly easy to count.

All these strange pollen grains do not come into function. Even

before the flower opens, they die, and the anthers then contain merely pulpy remains and resorbed pollen grains. When the flower opens, the anthers are empty, and no pollen will adhere to the numerous bees which all day suck honey from the multitude of flowers that adorn the Lyngby bog in the month of June.

It is true that, after the flowering, fruits develop in the other *Oxycoccus* species with normal pollen in the Lyngby bog. However, if we examine the large, luxuriant tufts of *O. gigas*, only few fruits will prove to continue their growth, the majority of them will soon stop growing and wither. But there always remain some which attain a considerable size; though they contain no embryo. I found embryo-sacs in the flowers examined; but the nucellus was almost entirely reduced. Germinated pollen (from the other species) was not observed by me either.

On the other hand, (Mycorrhiza?) fungi often penetrated down through the style, soon surrounding the barren seeds with a thick layer of hyphae.

Thus, as far as I have been able to observe, seed capable of germinating does not seem to develop in *O. gigas*. That it may nevertheless take place in exceptional cases is a hypothesis that cannot be entirely rejected.

Accordingly, the plant does not seem to have sexual reproduction; and how it is dispersed from place to place, is a mystery. However, the cytological conditions would seem to indicate that *O. gigas* is possibly a hybrid between a species with $n = 12$ and another with $n = 24$. If this is correct, it may have arisen several times and in different places, where it has then held its own and spread vegetatively. An argument against this supposition, however, is that at present no *Oxycoccus* with $n = 12$ is found, for instance, in the Lyngby bog nor in Denmark at all. But such a species (*O. microcarpus*) may very well have grown there in subglacial times, and its hybrid with the subsequently immigrated *O. quadripetalus* ($n = 24$) may have held its own up to the present day — thanks to the good ecological equipment which it possesses as a polyploid — in connection with its great power of vegetative propagation.

SUMMARY.

1) The genus *Oxycoccus* includes several closely related forms, whose chromosome numbers constitute a polyploid series, viz. $n = 12$, $n = 24$, and $n = 36$.

2) n = 12 in *O. microcarpus*, the species found farthest north, and which also differs ecologically from *O. quadripetalus*. Cf. further SAMUELSSON (1922).

3) n = 2 × 12. *O. quadripetalus* is the most commonly occurring species and is present in the greatest numbers. Var. *microphyllus* is its outpost to the north (Greenland).

4) n = 3 × 12. The newly discovered *O. gigas* (= *O. quadripetalus* × *O. microcarpus?*) is hexaploid. It does not produce germinable pollen, and is sexually sterile. For further details, see pp. 406—409.

5) Accordingly, the polyploid series dealt with here attains its optimum at tetraploidy. With higher chromosome numbers sexual reproduction ceases. The same thing applies to the genus *Deschampsia* (HAGERUP, 1939).

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DIE MEIOSIS BEI HAPLOIDEN PFLANZEN VON GODETIA WHITNEYI

VON *ARTUR HAKANSSON*

LUND

(With a Summary in English)

D AS Material zu vorliegender Untersuchung stammt von Dozent GUNNAR HIORTH, Landwirtschaftliche Hochschule in Ås, Norwegen, der es mir in freundlicher Weise zwecks zytologischer Untersuchung zur Verfügung gestellt hat. Es wurden ziemlich viele Mutationen untersucht, die aus Inzuchtlinien von *Godetia Whitneyi* wie auch aus einigen von *G. amoena* erhalten worden waren. Hier sollen die Ergebnisse meiner Studien über Haploide mitgeteilt werden, von denen in den Kulturen mehrere Pflanzen angetroffen worden sind.

Die untersuchten Blütenknospen sind in NAWASCHINs Flüssigkeit fixiert worden. Sie wurden eingebettet und am Mikrotom geschnitten. Die Färbung erfolgte mit HEIDENHEINs Eisenhämatoxylin. Die gemachten genetischen Angaben wurden mir von Dozent HIORTH freundlichst übermittelt. Meine *Godetia*-Untersuchungen sind mit Unterstützung des BOKELUND-Fonds der Universität Lund ausgeführt.

Zwei haploide Pflanzen wurden untersucht, G. 4359/220 (Fixierungsnr. 211) und G. 2930/132 (Fixierungsnr. 48). Erstere ist nach Bestäubung mit röntgenbehandeltem Pollen, letztere spontan entstanden. Ausserdem erwies sich eine *G. amoena*-Pflanze als haploid. Es war dies G. 8474/581 (Nr. 214), aber Meiosisstadien konnten hier nicht untersucht werden.

Die Meiosis ist an zahlreichen Haploiden studiert worden (siehe IVANOV, 1938). Ihr Verlauf wird von verschiedenen Forschern verschieden geschildert, was zum Teil auf das Material zurückzuführen ist, aber teilweise auch auf die Interpretation desselben, wie aus den Arbeiten über die Meiosis haploider *Oenothera* hervorgeht. Besonders schwierig ist es die Bewegungen der univalenten Chromosomen nach der Diakinese klarzulegen. Bei *Godetia* wird die Serienkombination der beobachteten Stadien in der Entwicklung dadurch erleichtert, dass die Pollenfächer in durch steriles Gewebe getrennte, kleinere Abteilungen aufgeteilt sind. Die PMZ in zwei angrenzenden Abteilungen

zeigen grössere Unterschiede in der Entwicklung als die PMZ in ein
und derselben Abteilung.

Diakinese. — Die Diakinese zeigt 7 Univalente, die deutlich längs-
gespalten sind (Fig. 1 *a*). Auch im zunächst vorhergehenden Stadium,
wo die Chromosomen nicht so stark kontrahiert sind, erkennt man die
Chromatiden sehr deutlich. Zwei Nukleolen sind bisweilen im Dia-
kinesekern.
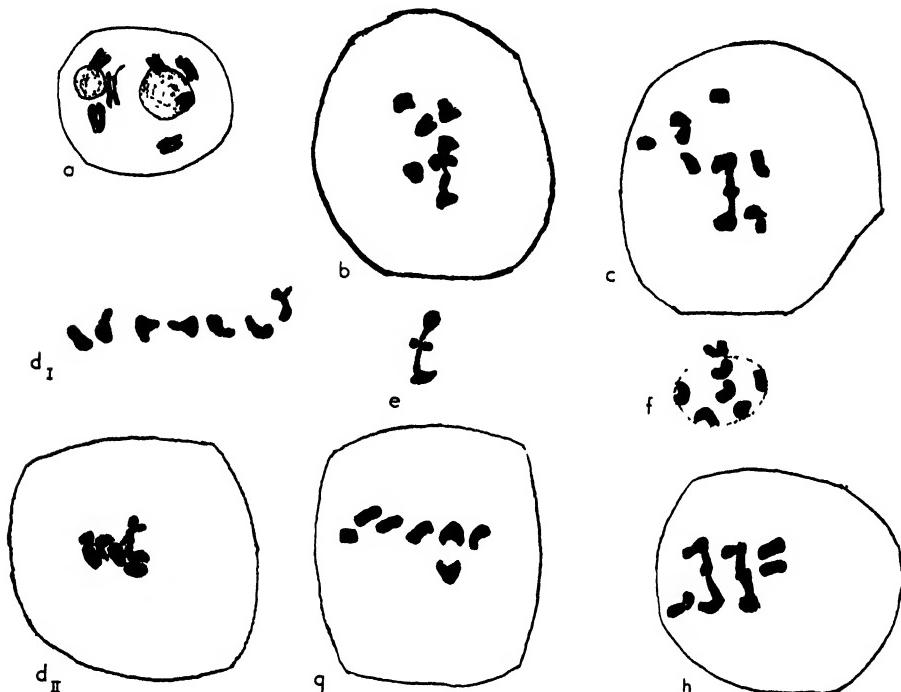
Fig. 1. — *a*: Diakinesekern. — *b*: PMZ mit Bivalent. — *c*: PMZ mit Bivalent und
fragmentiertem Chromosom. — *d*$_I$ und *d*$_{II}$: PMZ mit Kernplatte und die Chromo-
somen dieser Platte. — *e*: Bivalent mit subterminalem Chiasma. — *f*: Ein Chromo-
som ist ausserhalb der Spindel. — *g*: PMZ mit Ringbivalent. — *h*: PMZ mit zwei
Bivalenten.

Bivalente. — Kaum 1 % der PMZ enthalten ein Bivalent. Dieses
war stets in der Längsrichtung der Kernspindel normal orientiert. Es
hatte ein Chiasma, das gewöhnlich terminal (Fig. 1 *b*), seltener sub-
terminal (Fig. 1 *e*) gelegen ist. Ein einzigesmal wurden zwei Bivalente
beobachtet, von denen das eine deutlich heteromorph war (Fig. 1 *h*).
Im allgemeinen sind indessen die beiden gepaarten Chromosomen ein-
ander sehr ähnlich (andere Beispiele für Heteromorphie sind jedoch in
den Fig. 1 *e* und 3 *a* zu sehen). Einige wenige Male wurde ein ring-

förmiges Bivalent mit zwei terminalen Chiasmata beobachtet (Fig. 1 *g*). Dass die Frequenz der Bivalentbildung von ausserhalb der Chromosomen liegenden Faktoren beeinflusst wird, darauf deutet, dass während in den meisten Kleinfächern keine PMZ ein Bivalent aufweist, in anderen zuweilen eine, aber auch zwei oder drei PMZ ein solches enthalten. Einige Forscher haben der Ansicht Ausdruck gegeben, dass die bei Haploiden beobachteten Bivalente keine echten solchen seien, sondern auf der Verschmelzung von heterochromatischen Teilen oder anderem beruhten. Aber die Orientierung der Centromeren in der Kernspindel und das Vorkommen von subterminalen Chiasmata sprechen natürlich dafür, dass es sich um eine wirkliche Chromosomenpaarung handelt, die ihre Erklärung im Vorkommen von duplizierten Stücken im haploiden Genom bekommt.

Es ist bei den meisten untersuchten Haploiden eine Bivalentbildung konstatiert worden, auch in solchen mit der »Grundzahl». Der Prozent PMZ mit einem Bivalent ist bei verschiedenen Haploiden sehr ungleich. Auch innerhalb derselben Art kann er variieren, so bei *Oenothera franciscana*, wo in ein paar Knospen fast alle PMZ ein Bivalent hatten, während in einem anderen »most of the cells failed to show» ein Bivalent (EMERSON, 1929).

Metaphase und Anaphase. — Nach dem Verschwinden der Membran des Diakinesekerns liegen bei di- und polyploiden Godetien Bivalente und Multivalente etwas zerstreut, aber sie ordnen sich schnell zu einer Metaphaseplatte. Dieses Stadium währt lange Zeit, worauf die Anaphase schnell stattfindet. Zufällig vorkommende univalente Chromosomen sind entweder in der Platte oder näher dem einen Pol beobachtet worden. Bei den haploiden Godetien verhalten sich die seltenen Bivalente wie bei den Diploiden, aber die Anwesenheit von zahlreichen Univalenten gibt ein sehr variierendes Bild dieser Stadien.

Die Univalente sind ziemlich kontrahiert, sie haben demnach kein »somatisches» Aussehen. Zuweilen kann man jedoch die Stelle der primären Einschnürung sehen. Drei Chromosomen haben ein submedianes Centromer, zwei Chromosomen ein subterminales, sodass die beiden Schenkel sehr ungleich lang sind, während in zwei Chromosomen der eine Schenkel etwa doppelt so lang ist als der andere (Fig. 2 *e*). Die Längsspaltung der Univalente ist nicht mehr wahrzunehmen, sie wird erst später wieder deutlich. Einigemale wurden abweichende Chromosomen beobachtet. Fig. 1 *c* zeigt eine Zelle, in der ein Univalent beim Centromer zweigeteilt ist. Fig. 3 *d* zeigt eine Zelle mit einem extra Fragment. Sehr häufig liegen die Chromosomen

nicht in der Mitte der PMZ sondern neben der Wand. Solchenfalls
wird die Kernspindel später, wenn sie sich stark verlängert, gekrümmt,
etwa halbmondförmig, da sie nicht genügend Platz bekommt und wohl
auch an den gerundeten Wänden nicht genügend Widerstand findet,
als dass die Verlängerung verhindert und die Spindel gerade verbleiben
würde. Früher ist hervorgehoben worden, dass die Kernspindel bei
schwacher Chromosomenpaarung mit vielen Univalenten sich viel mehr
verlängert als wenn Bivalente vorhanden sind (siehe BEADLE, 1933).
Die Ausbildung der Kernspindel bei der haploiden *Godetia* ist aber sehr
verschieden. Schliesslich sei hervorgehoben, dass ein Univalent zu-
weilen ausserhalb der Kernspindel liegen kann. Solche Chromosomen
scheinen zu degenerieren; sie haben anscheinend kein Vermögen einen
Kern zu bilden. Fig. 1 *f* zeigt ein Chromosom ganz neben der Kern-
spindel (und der sich ausbildenden Kernmembran).

Es ist ziemlich schwierig während der recht langen Zeit, die vor
Beginn des Interkinesestadiums verfliesst, auf die Bewegungen der
Univalente in der Kernspindel zu schliessen. Nach Auflösung der Kern-
membran liegen die Univalente ein wenig zerstreut. In vielen PMZ
ordnen sich einige oder alle Univalente zu einer Kernplatte, aber in
anderen PMZ findet dies offenbar nicht statt. Fig. 1 *d* zeigt eine PMZ,
in der ziemlich unmittelbar nach dem Verschwinden der Kernmembran
eine Kernplatte gebildet worden ist, also ebenso schnell wie bei den
Diploiden. Dies ist indessen eine Ausnahme, die Regel ist, dass die
Univalente sich langsam und nach und nach äquatorial ordnen. In
den übrigen in Fig. 1 abgebildeten PMZ ist nach Auflösung der Kern-
membran sicher längere Zeit verflossen als in *d*; aber nur in *g* liegen
alle Chromosomen in der Kernplatte. Diese ist häufig ungleichmässig;
es hat den Anschein, als ob es den Univalenten schwer fiele sich richtig
einzustellen.

Häufig kommt es vor, dass sich keines der Chromosomen äqua-
torial ordnet. In Fig. 2 *a* bilden sie anstatt dessen eine sphärische
Gruppe, als ob sie von den Polen keinem Druck ausgesetzt wären; es
scheint nur die Repulsion zwischen den Centromeren der Chromosomen
zu wirken. In diesem Stadium erscheint dann wiederum die Längs-
spaltung (Fig. 2 *d*) und es wird ein kugelrunder Interkinesekern ge-
bildet (wie in Fig. 7 *a*). Restitutionskerne von diesem Aussehen waren
sehr häufig. Es ist zu beachten, dass die Spaltung nicht das Centromer
trifft; dieses verbleibt ungeteilt und hält die beiden Chromatiden zu-
sammen, die auseinander weichen. Chromosomen mit medianem
Centromer gleichen dann einem symmetrischen X. Die Chromosomen

können indessen in der PMZ auch zerstreuter liegen. Fig. 2 *b* zeigt ein Beispiel hierfür, obgleich es vielleicht nicht ausgeschlossen ist, dass es in dieser Zelle früher eine Kernplatte gegeben hat. In dieser PMZ wird dann ein grösserer und langgestreckter Interkinesekern gebildet (wie in Fig. 6 *a*).

Ein Univalent ordnet sich also nur mitunter in eine Kernplatte ein.

Fig. 2. — *a*: PMZ mit sphärischer Univalentengruppe. — *b*: PMZ mit mehr zerstreuten Univalenten. — *c*: Vier Univalente in der Kernplatte — *d*: Weiterentwicklung von *a*. — *e*: Die sieben Univalente aus einer Zelle. — *f*: Fünf Univalente in der Kernplatte. — *g*: Univalente auf der Kernspindel zerstreut. — *h*: Drei Univalentengruppen in der PMZ. — *i*: Anaphase mit der Verteilung 3 + 4.

Ist es einmal dahin gekommen, kann es sich indessen auf zwei verschiedene Weisen verhalten: entweder verbleibt es in der Kernplatte oder auch es unternimmt eine Anaphasewanderung nach einem der Pole. Aber, wie sich die Chromosomen auch verhalten mögen, so verbleibt ihr Centromer meistens ungeteilt, also auch wenn es in der Kernplatte liegen bleibt. Zufolge dieser Variation im Verhalten der Univalente entstehen viele verschiedene zytologische Bilder, und nicht wenige von

diesen können nicht mit Sicherheit gedeutet werden. In bezug auf die
in Fig. 2 c abgebildete PMZ, wo vier Chromosomen eine Kernplatte
bilden, lässt sich unmöglich entscheiden, wie sich die ausserhalb der
Platte liegenden Univalente verhalten werden. In dem oberhalb der
Platte liegenden ist das Centromer gegen die Platte gerichtet, in dem
einen der beiden unterhalb der Platte gelegenen Chromosomen ist es
von der Platte gerichtet, und im zweiten sind die Chromosomenarme
longitudinal orientiert. Die Centromeren würden am ehesten darauf
deuten, dass sich alle drei Chromosomen verschieden verhalten, aber
es erscheint fast absurd, dass ein Chromosom sich am Weg zur, das
zweite von der Platte befindet und das dritte stationär sein sollte;
wahrscheinlicher sind alle drei stationär und die Orientierung der
Centromeren dürfte eine zufällige sein. Eine andere schwer zu er-
klärende PMZ zeigt Fig. 2 f. Fünf Univalente bilden eine Platte, die
nicht äquatorial sondern näher dem einen Pol in der PMZ liegt (die
Umgrenzung der Kernspindel war, wie bei Haploiden häufig, undeut-
lich, wahrscheinlich ist sie oft in irgend einer Weise abnorm), darunter
liegen zwei Chromosomen, die ein Bivalent zu bilden scheinen.

Fig. 3 b zeigt dagegen unzweideutig eine Polwanderung der Uni-
valente. Hier sind, wie es scheint, alle Centromeren in jeder Gruppe in
der Richtung vom Äquator orientiert. Das oberste Chromosom war bei
einer anderen Einstellung der Mikrometerschraube mehr sichtbar als
die übrigen, weshalb es wahrscheinlich erscheint, dass es niemals in der
Äquatorialplatte gelegen hat. Die Befestigung von Zugfasern am Chro-
mosom wurde beobachtet. Fig. 2 i zeigt eine 3 + 4 Verteilung; es
scheint, als ob es hier ein Bivalent gegeben habe, dessen Chromosomen
auf zwei Pole verteilt werden. Bivalente sind in diesem Material so
selten, dass es nicht möglich ist zu sagen, ob dies ihr gewöhnliches
Verhalten ist. Auch andere Verteilungen wurden beobachtet. Es ist
sehr häufig, dass ein Chromosom am einen Pol, sechs beim anderen
liegen (eine Verteilung, die bei vielen anderen Haploiden angetroffen
worden ist). Häufig beruht dies darauf, dass das einzelne Chromosom
von Beginn an dem einen Pol näher lag und bei der Streckung der
Spindel diesem näher gebracht worden ist. Es hat aber tatsächlich
den Anschein, als ob die Anaphasenverteilung 6 + 1 einer Kernplatte
häufiger auftritt, als man auf Grund der Zufallsgesetze erwarten würde.
In den Fig. 3 c und d sehen die Chromosomen der einen Polgruppe
ganz so aus wie in einer normalen diploiden Gruppe. Bei der Ana-
phasenwanderung resultieren zwei Interkinesekerne wenn die beiden
Chromosomengruppen gut getrennt sind. Oft sind jedoch die Chromo-

somen längs der Kernspindel zerstreut (Fig. 2 g), und dann entsteht ein
einziger, aber langer und schmaler Interkinesekern.

Häufig gibt es auf der Kernspindel mehr als zwei Chromosomen-
gruppen. Fig. 2 h zeigt am unteren Pol eine, am oberen Pol zwei
Chromosomen, während vier Chromosomen zunächst eine unregelmäs-
sige Kernplatte bilden. Fig. 3 a zeigt ein Chromosom an jedem Pol,
während die übrigen nahe des Äquators liegen. Andere Beispiele für
drei Gruppen sind Fig. 4 f und g. Es werden auch, wenngleich selten,

Fig. 3. — a: Drei Chromsomengruppen auf der Spindel. — b: Anaphase 3 + 4. —
c: Anaphase 6 + 1. — d: Späte Anaphase 6 + 1. — e: PMZ mit Fragment. — f: PMZ
mit Kernplatte und zwei isolierten Chromosomen. — g: Quergeschnittene Univalenten-
platte. — h: Ditto. Weiterentwicklung von g. — i: Anaphasechromosom mit
Zugfasern.

drei Interkinesekerne gebildet (Fig. 6 d). Die in Fig. 6 g abgebildete
Lage der drei Kerne ist vielleicht infolge einer starken Streckung einer
peripher liegenden Kernspindel entstanden, wobei ein bzw. zwei Chro-
mosomen, die nahe den Polen gelegen sind, weit weg geschoben wor-
den sind.

Wir kommen schliesslich zu dem Fall, in dem die Univalente sich
in eine regelmässige Platte geordnet haben, diese aber dann nicht ver-
lassen. Fig. 3 f ist etwas ungewöhnlich. In der gleichen Ebene liegt

eine Kernplatte mit fünf Chromosomen und jederseits dieser ein einzelnes Chromosom. Letztere liegen ausserhalb der Spindel. In Fig. 3 g hat die PMZ eine Platte mit 7 Chromosomen, von denen einige eine Andeutung zu Teilung aufweisen; Fig. 3 h zeigt auch eine Kernplatte von der Polseite; aber hier haben die Chromosomen begonnen Interkineseform anzunehmen. Fig. 4 a zeigt eine solche Kernplatte von der Seite; um eines der Chromosomen gibt es eine Andeutung zu Mem-
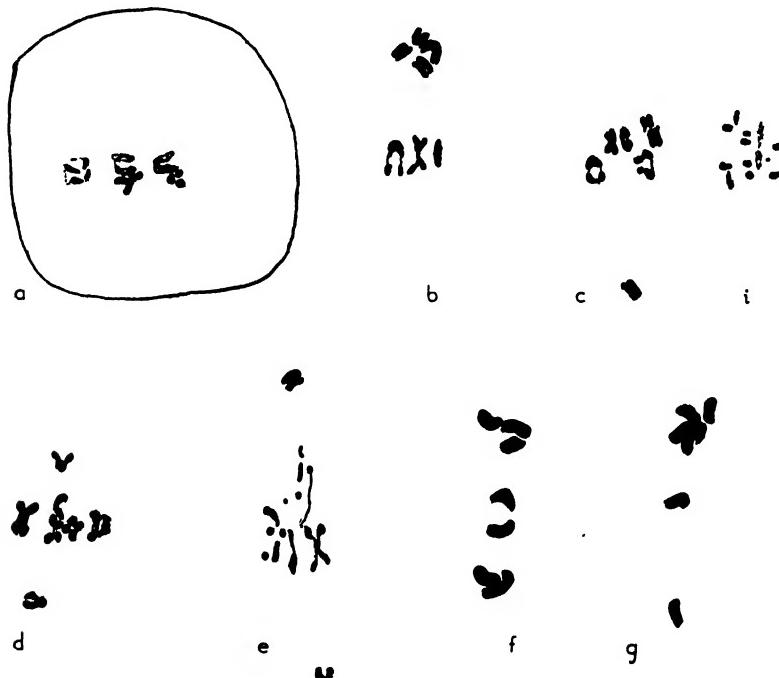
Fig. 4. — a: PMZ mit Kernplatte, die Univalente nehmen Interkineseform an. — b: Vier Univalente bei dem Pol, drei in der Platte. — c: Sechs Univalente in der Platte. — d: Fünf Univalente in der Platte. — e: Weiterentwicklung aus a—d, die Plattenunivalente scheinen sehr langgestreckt und teilweise fragmentiert zu sein. — f und g: Drei Chromosomengruppen auf der Spindel. — i: Teilung der Univalente (?).

bran. Die Chromosomen haben auch das gleiche Aussehen wie beim Übergang zu gewöhnlicher Interkinese (vgl. Fig. 5). Oft entsteht grosse Ähnlichkeit mit dem Beginn der Metaphase 2; dass es sich aber nicht um dieses Stadium handelt, ergibt sich bei einem Vergleich mit den anderen PMZ im Kleinfach und aus den Fällen, in denen nur ein Teil der Chromosomen in der Platte liegt. Fig. 4 b zeigt vier Chromosomen am einen Pol, während nur drei im Äquator liegen. In Fig. 4 c liegen dagegen·sechs in der Kernplatte, und in Fig. 4 d schliesslich fünf,

während hier ein Chromosom an jedem Pol liegt. Man würde nun erwarten, dass sich die Chromatiden trennten und eine Längsteilung wie bei einer pseudohomotypischen Teilung einträte. Aber ein sicherer solcher Fall ist nicht beobachtet worden. Die Chromosomen in der Platte sehen oft wie kleine Bivalente aus, ihre Chromatiden sind stark ausgespannt, als ob sie von verschiedenen Richtungen einem Zug ausgesetzt seien, aber der Kontakt zwischen ihnen scheint im allgemeinen nicht verloren zu gehen. Es hat den Anschein, als ob die Kernspindel sich verlängert und das Centromer sich am Nullpunkt befindet. Oder es kann aussehen, als ob das Centromer geteilt ist, die Tochtercentromeren aber durch einen Faden verbunden sind. Schliesslich bekommt man mitunter den Eindruck, als ob das Chromosom eine Querteilung statt einer Längsteilung erfahren sollte (»misdivision of centromere»; DARLINGTON, 1939). Die Chromosomenarme sind nämlich oft in der Längsachsel der Spindel (Fig. 2 h). Die am Äquator liegenden Chromosomen bekommen hierdurch ein anderes Aussehen als die an den Polen liegenden (Fig. 4 e). Die zunächst folgende Entwicklung erschien schlecht fixiert. Sowohl bei 48 wie bei 211 waren die aus der Kernplatte gebildeten Interkinesekerne äusserst undeutlich. Sie waren wenig färbbar, und was man von den Chromosomen sehen konnte waren lange Schleifen oder mehr oder weniger zahlreiche Fragmente (noch undeutlicher als in Fig. 4 e abgebildet). Fig. 4 i zeigt vielleicht eine Teilung der Univalente.

Bei den *Godetia Whitneyi*-Haploiden kann demnach von einer »zufallsweisen» Verteilung der Univalente auf die beiden Pole keine Rede sein. In den allermeisten PMZ wird ein einziger Interkinesekern mit sieben Chromosomen gebildet. Und wenn zwei Kerne gebildet werden, was in kaum mehr als $^1/_4$ der PMZ vorkommt, so ist es das häufigste, dass der eine nur ein Chromosom enthält (also die »Verteilung 6 + 1»). Aber eine Verteilung von 2 + 5 und 3 + 4 wurde natürlich verschiedenemale beobachtet; sie entsteht am häufigsten durch eine Polwanderung von einer Kernplatte. Wie wenn es sich um die Bivalentbildung handelte, konnte man übrigens auch beim Studium der Meta-Anaphase konstatieren, dass bei den im gleichen Kleinfach gelegenen PMZ eine Tendenz bestand, sich gleichartig zu verhalten. So hatten fast alle PMZ in einem Kleinfach von Pflanze 48 eine Gruppe von sechs Chromosomen und ein einzelnes Chromosom. In einem anderen Kleinfach zeigten dagegen 11 PMZ »7 + 0» und 2 PMZ »6 + 1».

Beim Übergang zur Interkinese ist die Längsspaltung in den Chro-

mosomen wiederum sichtbar, wobei jedoch das Centromer ungeteilt
verbleibt. Fig. 3 c zeigt eine Anaphase mit noch ungeteilten Chromo-
somen, die Chromosomenverteilung scheint 6 + 1 zu werden. Fig. 3 d
hat die gleiche Verteilung in einem etwas späteren Stadium; hier ist die
Spaltung deutlich. In Fig. 5 a sind die Chromosomen beim Eintritt in
das Interkinesestadium längs einer peripheren Kernspindel zerstreut,
Fig. 5 b ist ähnlich, aber es scheinen zwei Kerne nebeneinander gebildet
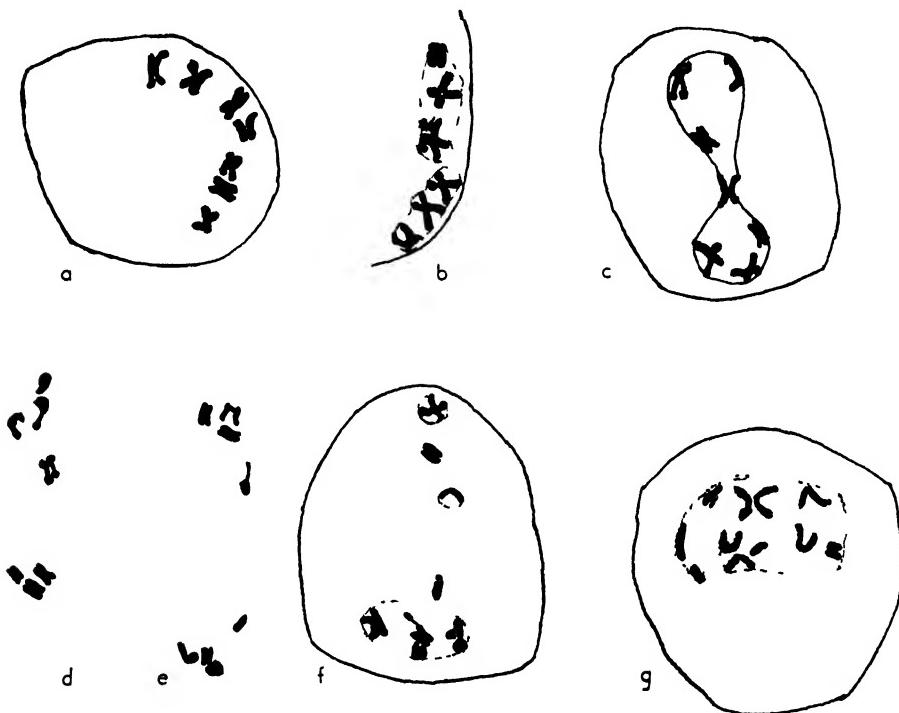
Fig. 5. — a—c: Übergang zur Interkinese bei verschiedener Lage der Univalenten. —
d: Anaphase, ein Univalent verspätet. — e: Das Centromer in dem verspäteten Uni-
valent ist geteilt. — f: Interkinese, ein Centromer ist geteilt — g: Einige Centromere
in dem Interkinesekern sind geteilt.

zu werden, die wahrscheinlich bald verschmelzen. Die Kernbildung
beginnt rund um jedes Chromosom, und wenn diese getrennt liegen,
tritt gelegentlich das Bild von Kleinkernen auf, die dann ineinander
übergehen. In Fig. 5 c ist ein stundenglasförmiger Interkinesekern
gebildet worden.

   Zuweilen kommt es wirklich zu einer Teilung des Centromers.
Dies kann in einem in der Kernspindel verzögerten Chromosom vor-
kommen (was auch bei untersuchten di-polyploiden Typen beobachtet

worden ist). Fig. 5 *d* zeigt drei Chromosomen an jedem Pol, während
ein verspätetes Univalent sich in Teilung befindet. Fig. 5 *e* ist ähnlich,
aber die beiden Längshälften sind weit getrennt. Fig. 5 *f* zeigt eine
unregelmässigere Verteilung. Am oberen Pol liegen zwei Chromoso-
men, von denen das eine sich zu einem Kleinkern ausgebildet hat; am
unteren Pol gehören vier Chromosomen zu einem Kern, dessen Mem-
bran noch nicht fertig ist. In der Mitte der PMZ gibt es zwei
Chromatiden, deren Umbiegung zeigt, dass das Centromer in einem
Univalent geteilt worden ist, und dass darauf eine Repulsion der Toch-
tercentromeren stattgefunden hat. Das obere dieser Chromatiden hat
einen eigenen Kleinkern gebildet, in bezug auf das untere ist es un-
gewiss, ob es in den grossen Kern eingehen wird. Fig. 5 *g* zeigt einen
Interkinesekern, in dem die Centromeren in einigen der Chromosomen
geteilt sind; am deutlichsten in bezug auf die beiden V-förmigen
Chromatiden rechts im Kern.

Interkinese. — Im Interkinesekern wurden einige Beobachtungen
über Nukleolen gemacht. Seit HEITZ ist nachgewiesen, dass Nukleolen
an bestimmten Chromosomen, so besonders an SAT-Chromosomen,
gebildet werden. In den Fällen wo SAT-Chromosomen fehlen, gleich-
wie auch achromatische Einschnürungen an anderen Chromosomen,
werden Nukleolen an den Enden gewisser Chromosomen gebildet (siehe
GEITLER, 1938, S. 29 und MATSUURA, 1939). Da ich keine guten somati-
schen Teilungen zu studieren Gelegenheit hatte, kann ich mich über das
ev. Vorkommen von achromatischen Teilen in den *Godetia*-Chromo-
somen nicht äussern; aber wahrscheinlich gibt es SAT-Chromosomen.
Es zeigt sich nämlich, dass in der Interkinese zwei Chromosomen
»nukleolenbildend» sind. Das eine dieser Chromosomen hat ein sub-
terminales Centromer und einen Nukleolus am einen Ende, wo sich
wahrscheinlich ein Satellit befindet, wenngleich er in diesem Stadium
undeutlich zutagetritt (siehe Fig. 6 *b* u. *f*). Das zweite hat den Nukleolus
submedian (siehe Fig. 6 *e*).

Das Chromosom, das den Nukleolus terminal bildet, bekommt
eigentümlicherweise gern Zwillingsnukleolen von ganz gleicher Grösse
(Fig. 6 *e* und *g*). Diese scheinen später zu einem vereinigt worden zu
sein (Fig. 7 *a*). Es ist schon hervorgehoben worden, dass die Chromo-
somen in diesem Stadium längsgespalten sind; das Vorkommen von
Zwillingsnukleolen zeigt, dass der nukleolenbildende Teil des Chromo-
soms auch doppelt ist. Die Grösse des Nukleolus ist vom Chromosom
abhängig; es zeigt sich ja, dass zwei identische nukleolenbildende Teile
auch gleich grosse Nukleolen bilden.

Wenn die PMZ einen Interkinesekern enthält, hat er in den meisten Fällen nur einen einzigen Nukleolus, und dieser pflegt an dem Chromosom befestigt zu sein, das den Nukleolus terminal ausbildet; dieses ist also am wichtigsten, es hat das grösste nukleolenbildende Vermögen (Fig. 6 g, 7 a, während Fig. 6 a, wie es scheint, ein anderes Verhalten aufweist). Seltener bilden beide Chromosomen im Kern einen Nukleolus (Fig. 6 e). Fig. 6 b zeigt eine Interkinese mit der
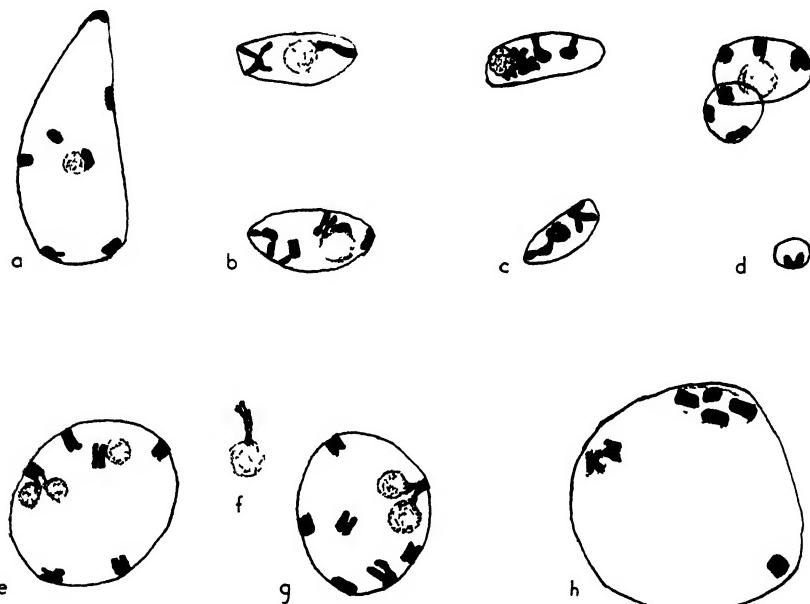
Fig. 6. — a: Interkinesekern mit einem Nukleolus — b und c: die »nukleolenbilden-den» Chromosomen sind auf zwei Kerne verteilt. — d: Sie sind wahrscheinlich beide in dem Interkinesekern mit vier Chromosomen. — e: Beide haben Nukleolen gebildet, das »terminale» Zwillingsnukleolen, trotzdem sie sich in demselben Kerne befanden. — f: Das »terminale» Nukleolenchromosom. — g: Kern mit Zwillings-nukleolen. — h: PMZ mit drei Interkinesekernen.

Chromosomenverteilung 5 + 2 und die beiden nukleolenbildenden Chromosomen liegen in verschiedenen Kernen. Ähnlich ist Fig. 6 c. Die Abwesenheit einer Konkurrenz vom »terminalen Chromosom» gibt dem anderen Gelegenheit einen Nukleolus auszubilden. In Fig. 8 a haben dagegen die beiden Pollenzellen mit zwei Chromosomen keinen Nukleolus, und dies dürfte wohl darauf beruhen, dass beide nukleolen-bildende Chromosomen in den 5-chromosomigen Interkinesekern ein-gegangen sind. Von den drei Interkinesekernen in Fig. 6 d enthält nur einer, der mit vier Chromosomen, einen Nukleolus, was wahrschein-

lich auf das Gleiche beruht, dass nämlich beide Nukleolenchromosomen in den gleichen Kern gelangt sind.

MATSUURA (1939) hat gezeigt, dass es bei *Trillium kamtschaticum* zwei nukleolenbildende Chromosomen gibt (die Nukleolen werden am einen Ende von Chromosom *A* und *E* gebildet). Bei asynaptischen
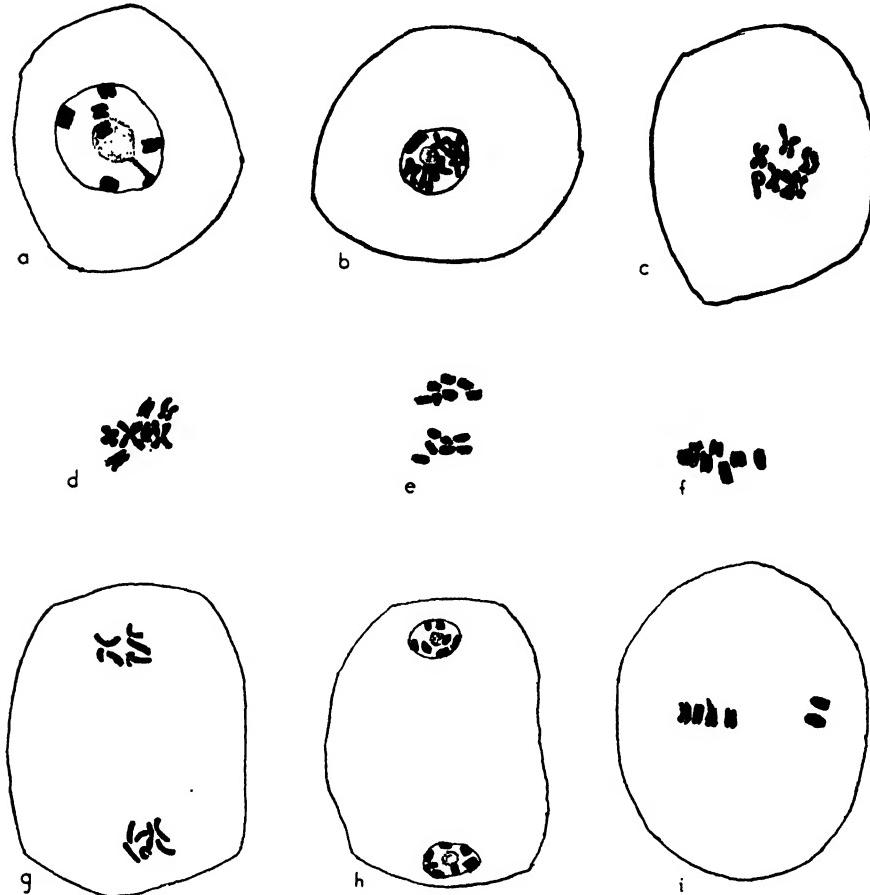
Fig. 7. Die zweite Teilung. — *a* und *b*: Interkinese. — *c* und *d*: Übergang zur Metaphase 2. — *e*: Anaphase 2. — *f*: Metaphase 2. — *g*: Späte Anaphase. — *h*: Die Pollenkerne sind gebildet. — *i*: PMZ mit zwei Kernspindeln.

Pflanzen, wo die Chromosomen in der Meiosis isoliert bleiben und verschiedene Kleinkerne bilden, zeigte es sich, dass alle das Vermögen zur Bildung eines Nukleolus haben. Dieses Vermögen ist bei *A* und *E* nur viel grösser, sodass die Anwesenheit eines dieser in einem Kern die anderen Chromosomen an der Bildung eines Nukleolus hindert. Aber

es scheint, als ob haploide *Godetia* sich in anderer Weise verhielten. Man findet häufig Kerne mit einem oder einigen wenigen Chromosomen ohne deutlichen Nukleolus. Alle Chromosomen scheinen hier kaum das Vermögen zur Bildung von Nukleolen zu besitzen.

Die zweite Teilung. — Diese wurde nicht so eingehend studiert, wie erwünscht war, da sie recht schlecht fixiert war. Fig. 7 *a*—*h* zeigt den Verlauf der Teilung eines Interkinesekerns mit 7 Chromosomen. Er erfährt zuerst gern eine Kontraktion (Fig. 7 *b*), Kernmembran und Nukleolus werden aufgelöst, und die Chromosomen, deren Chromatiden stark auseinander weichen, aber von den Centromeren zusammengehalten werden, liegen zuerst ein wenig unregelmässig (Fig. 7 *c* und *d*), ordnen sich aber dann zu einer Kernplatte (Fig. 7 *f*). Wenn die Metaphase ihren Höhepunkt erreicht, sind die Chromosomen stärker kontrahiert und nicht mehr X-ähnlich (Fig. 7 *f*, vgl. 7 *i*). Die Anaphase ist in Fig. 7 *e* und *g* abgebildet, die Schwesterchromatiden trennen sich. In den 7-chromosomigen Telophasenkernen wird der Nukleolus am einen Chromosom gebildet (Fig. 7 *h*). Schliesslich wird eine Pollendiade gebildet (Fig. 8 *a*).

Wenn es mehrere Interkinesekerne gibt, werden sie alle geteilt (Fig. 7 *i*). Sie konnten nicht näher studiert werden. Wenn der eine Interkinesekern nur ein Chromosom hatte, wurde seine Membran jedoch aufgelöst und er teilte sich; aber es hatte den Anschein, als ob er während dieser Teilung zuweilen in die grössere Kernteilungsfigur einverleibt wurde. Solchenfalls sollte aus zwei Interkinesekernen mit 6 und 1 Chromosomen anstatt einer Tetrade mit zwei Zwergzellen eine Diade entstehen. Nicht selten wurde nur eine Zwergzelle beobachtet. Wahrscheinlich haben sich die Chromatiden nicht getrennt. Zurückgebliebene Chromosomen wurden während der zweiten Teilung beobachtet. Es handelt sich, wie aus anderem *Godetia*-Material hervorgeht, um Chromatiden von Chromosomen, deren Centromeren früher geteilt worden sind.

Sporaden. — Als Folge der Meiosis resultiert eine grosse Anzahl von Pollendiaden, obgleich es offenbar ist, dass der % in verschiedenen Kleinfächern beträchtlich variiert. Ferner gibt es Tetraden mit verschieden grossen Zellen (siehe Fig. 8 *a*), Triaden, deren Bildungsweise nicht klargelegt werden konnte, was auch für vereinzelte Monaden gilt. Auch Pentaden wurden, wenngleich selten, beobachtet (siehe Fig. 8 *b*). Bei einer Auszählung waren von 182 Sporaden 129 Diaden, also 71 %. Ein grosser Teil des Pollens bekam also trotz der Haploidie die normale Chromosomenzahl. Ferner gab es 28 Tetraden, also 15 %, 11

Triaden und 2 Pentaden.   Trotz des Verlaufes der Meiosis ist diese
Haploide indessen nur schwach fertil.   Bei Selbstung von 48 wurden
keine Nachkommen erhalten.   Dass aber befruchtungsfähige Pollen-
körner gebildet wurden, zeigte deutlich diploides ♀ ✕ haploides ♂,
welche Kreuzung als Resultat 28 Pflanzen lieferte.

Schliesslich sei erwähnt, dass bei der haploiden Pflanze von *Godetia
amoena* die Meiosis in anderer Weise verlaufen ist als bei *Whitneyi*.
Teilungsstadien gab es im fixierten Material nicht, aber die Frequenz
von Diaden unter den Sporaden war, auf Grund der recht fragmentari-
schen Studien zu urteilen, niedrig.   So wurden in einer Anthere, die
teilweise Sporaden enthielt, 4 Diaden, 4 Triaden, 8 Tetraden, 6 Pentaden
und 6 Hexaden gezählt.   Es ist klar, dass hier die Univalente mehr
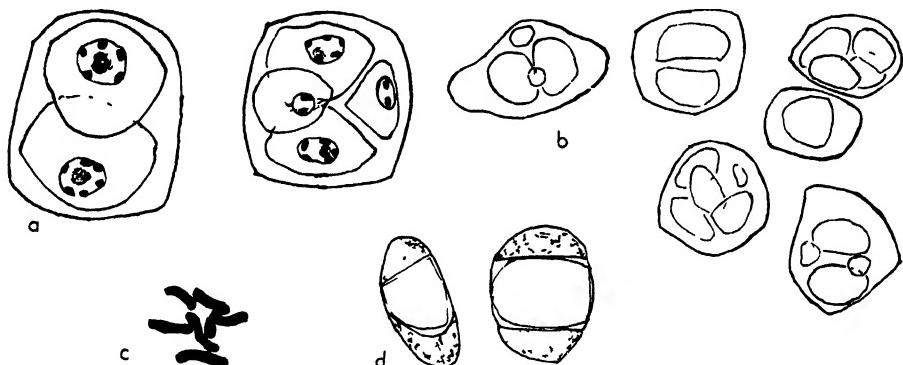
Fig 8   *a* und *b* Sporaden — *c G amoena* Somatische Kernplatte —
              d G amoena. Pollenkorner

zerstreut gewesen sind, sodass nur selten ein, dagegen oft drei Inter-
kinesekerne gebildet worden sind.   Die Chromosomenzahl wurde in
somatischen Platten bestimmt.   Fig. 8 *c* ist von einer Tapetumzelle.
Diese pflegen in der Regel viele Chromosomen zu enthalten, aber mit-
unter bleibt die Chromosomenvermehrung aus.

Die Pollenkörner der haploiden *amoena* waren von sehr variieren-
der Grösse und Form.   Zwei sind in Fig. 8 *d* abgebildet.   Indessen
scheint Nr. 214 laut Angaben von Dozent HIORTH fertiler zu sein als
haploide *Whitneyi*.   Bei Selbstung wurde eine geringe Anzahl Samen
erhalten, bei spontaner Kreuzung eine viel grössere Anzahl.   Die Eizellen
sind hier vielleicht besser als bei haploider *Whitneyi*.

Die ausserordentlich hohe Prozentzahl von unreduzierten Pollen-
körnern ist überraschend.   Pollendiaden sind bei vielen Haploiden fest-

gestellt worden, und auch dass solche aus unreduzierten Zellen bestehen können, aber der Prozent PMZ mit ausgebliebener Reduktion ist gering und schwankend, da er von äusseren Faktoren beeinflusst zu werden scheint. Ein recht hoher % wurde bei haploider *Datura stramonium* gefunden; nach Auszählung von Diaden wurden in einem Fall 10 %, aber in einem anderen 29 % Non-Reduktion berechnet (BELLING und BLAKESLEE, 1927); in einer diploiden Form, die durch ein rezessives Gen bedingte vollständige Asynapsis hatte, gab es dagegen nur 0—8 % Non-Reduktion (BERGNER, CARTLEDGE und BLAKESLEE, 1934). Nur bei einer haploiden *Matthiola incana* mit 7 Chromosomen und einem Fragment wurde eine ähnliche Prozentzahl wie bei *Godetia*, nämlich 70, gefunden, obgleich hier der Mechanismus, der zur Bildung der Diaden führte, ein anderer war (LESLEY und FROST, 1928).

Bei haploider *Whitneyi* wird das Centromer der Univalente während der Meiosis einmal geteilt, und dies ziemlich spät, sodass es bei Eintritt der Interkinese noch nicht stattgefunden hat. Nur selten erfolgt die Teilung früher und dann in einem verspäteten Anaphasechromosom, dessen Chromatiden dann häufig Zwergkerne bilden; diese können aber auch in einen grösseren Interkinesekern aufgehen. In der Regel liegen die Univalente so nahe aneinander, dass nur ein Interkinesekern gebildet wird. Dies beruht darauf, dass sich die Kernspindel nicht nennenswert gestreckt hat oder dass die Chromosomen bei der Streckung sehr zerstreut liegen. Eine variierende Anzahl von Univalenten bildet eventuell eine Kernplatte, sie sind, wie man sich auszudrücken pflegt, bipolar geworden, sie werden von den beiden Polen in der Kernspindel repelliert. Dieser bipolare Zustand geht bisweilen nicht verloren, sondern der Interkinesekern wird um die Äquatorchromosomen gebildet, die hierbei gern ein eigentümliches Aussehen annehmen (teilweise ähnlich dem von Pflanze B 7 von asynaptischem *Pisum sativum*; siehe KOLLER, 1938). In der Regel werden auch in diesem Fall die Centromeren nicht geteilt. Dagegen ist offenbar, dass die Univalente die Kernplatte verlassen und nach einem der Pole wandern können. Überhaupt verhalten sie sich wie eventuell vorkommende Univalente von diploiden *G. Whitneyi*-Formen (siehe HÅKANSSON, 1940).

Wenn haploide Pflanzen unreduzierte Pollenkörner bilden, kann dies, wie bei *G. Whitneyi*, durch die Bildung von Restitutionskernen geschehen, indem die Centromeren der Chromosomen bis zur zweiten Teilung ungeteilt verbleiben. Es resultiert eine Pollendiade. In anderen Fällen kommt es zu einer (laut GUSTAFSSON, 1935) pseudohomotypischen Teilung. Die Univalente bilden eine Kernplatte, ihre Centro-

meren teilen sich in dieser und es werden zwei Tochterkerne gebildet.
Eine neue Teilung findet nicht statt, sondern auch hier resultiert eine
Pollendiade.

Der erste Weg ist wohl der häufigste. Bei haploidem *Triticum
durum coerulescens* können wie bei *Godetia* ein oder mehrere Inter-
kinesekerne gebildet werden, abhängig von der Lage der Chromosomen
in der Kernspindel (KIHARA, 1936). Laut KIHARA liegen bei *Triticum*-
Bastarden eventuelle Univalente in früher Metaphase im allgemeinen
an den Polen, um darauf hinunter in die Äquatorialregion zu wandern
und eine Kernplatte zu bilden. Bivalente ordnen sich dagegen früh in
die Platte ein. Wenn nun bei in Frage stehenden Haploiden die Uni-
valente in die Interkinese übergehen, während sie sich an den Polen be-
finden, so werden zwei Interkinesekerne (eine Regressionsdiade) gebil-
det. Erfolgt dies später, wenn alle oder ein Teil von ihnen den Äquator
erreicht haben, so wird ein Kern (eine Regressionsmonade) von sehr
variierender Form gebildet. Die Entstehung einer Regressionsdiade als
Folge von Anaphasenwanderung der Univalente von der Platte nach
den Polen kommt also hier nicht vor, dagegen aber sicherlich bei der
haploiden *G. Whitneyi*. Ein Restitutionskern wird zuweilen bei ha-
ploiden *Oenothera blandina* gebildet (CATCHESIDE, 1932). Hier sollen
sich die Univalente früh in eine Kernplatte einordnen. Später erfolgt
die Polwanderung der Univalente, und wenn diese sehr zerstreut liegen,
kann ein einziger Interkinesekern gebildet werden.

Laut BLEIER (1933) ordnen sich dagegen bei haploiden *Oe. fran-
ciscana* und *Hookeri* die Univalente erst spät in eine Kernplatte ein. Die
Bildung von zwei Kernen erfolgt also bei diesen wie bei *Triticum* und
nicht wie bei *Oe. blandina*. Die Univalente, die die Kernplatte erreicht
haben, erleiden eine Längsteilung und die »Spalthälften» werden ent-
weder auf zwei Pole verteilt oder sie gelangen in einen einzigen Kern.
Es geht nicht richtig klar hervor, ob eine Teilung der Centromeren
immer stattgefunden hat, aber es erscheint wahrscheinlich, dass hier
der Restitutionskern von der Kernplatte gebildet werden kann. BLEIER
ist ferner der Ansicht, dass die Chromosomen in beiden Teilungen geteilt
werden können, dass also die Centromeren zweimal geteilt werden,
aber andere Forscher haben dies nicht gefunden. Bei diesen beiden
Haploiden sollten demnach laut BLEIER beide Typen und überdies
doppelte Teilungen der Centromeren vorkommen können. Bei haploider
G. Whitneyi kann vielleicht zuweilen auch eine pseudohomotypische
Teilung vorkommen, was jedoch nicht sicher festgestellt worden ist;
aber die Erfahrungen von anderen *Godetia*-Formen zeigen, dass das

Centromer nur einmal geteilt wird (HÅKANSSON, 1940), nach der pseudo-homotypischen Teilung also eine Pollendiade gebildet wird.

Ein typisches Beispiel für pseudohomotypische Teilung haben wir bei der früher erwähnten haploiden *Matthiola incana*. Hier bilden die Univalente eine Kernplatte, erfahren eine· Längsteilung, worauf zwei Tochtergruppen gebildet werden. Die zweite Teilung unterbleibt und es entsteht eine Pollendiade. Nur in einer geringeren Anzahl von PMZ haben sich die Chromosomen nicht geteilt, sondern wurden nach dem Zufall auf die zwei Pole verteilt. In diesen PMZ fand dann die zweite Teilung statt und es wurden Triaden und Tetraden gebildet. Dies scheint das beste Beispiel für eine pseudohomotypische Teilung bei Haploiden zu sein. Aus den Beschreibungen von haploiden *Datura stramonium* könnte man zur Auffassung gelangen, dass diese Teilung hier stattfindet, aber eine spätere Abhandlung über eine asynaptische diploide *Datura* gibt an, dass die Univalente sich dort wie bei den ha-ploiden verhalten und diese diploide Form bildet Restitutionskerne. Bei haploiden *Crepis capillaris* sollen die Univalente in seltenen Fällen in der Anaphase 1 geteilt werden (HOLLINGSHEAD, 1930). Einwände gegen die Deutung der Stadien in dieser Arbeit sind jedoch vorgebracht worden (BLEIER, l. c. S. 157).

SUMMARY.

Meiosis in two haploid plants of *Godetia Whitneyi* was studied.

Bivalents occur in less than 1 % of the pmc. The bivalents have a terminal or a subterminal chiasma. A few pmc had a ring bivalent. The bivalents indicate duplications in the haploid chromosome set.

In many pmc no metaphase plate is formed, in other pmc all or some of the univalents form a plate.

The univalents in the plate may go to the poles or remain in the plate.

Only very seldom does a centromere divide before interkinesis.

In most pmc only one interkinesis nucleus is formed. If there are two or three well separated groups of chromosomes two or three inter-kinesis nuclei are formed.

In interkinesis nuclei two chromosomes are able to form a nucleolus. One has the nucleolus terminally and may form twin nucleoli, the other has the nucleolus submedially. The former is the more important, because when both chromosomes are in the same nucleus, in most cases only this one has a nucleolus.

The second division proceeds normally, the centromeres splitting. About 70 % pollen diads with the unreduced chromosome number are formed.

A haploid *G. amoena* had few diads but pentads and hexads.

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CHROMOSOME NUMBERS AND POLY-PLOIDY WITHIN THE FLORA OF SPITZ-BERGEN

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THE extensive investigations of recent years on both experimentally produced polyploids and polyploids in nature have led to results which have greatly increased our knowledge of the evolutionary methods of nature. There can hardly be any doubt at present that polyploidy (the term including both autopolyploidy and allopolyploidy) has played a very prominent evolutionary role.

One of the problems to which many investigators have devoted their attention is the adaption of polyploids to extreme conditions of life. In this connexion it is of interest to call attention to the fact that in those cases in which both diploid and polyploid »races» occur within a Linnean species it has frequently been found that the polyploids have a more northern or alpine habitat than the diploids. As a classic example of this we may mention *Biscutella laevigata* (MANTON, 1934). The polyploid forms of this species have their habitat in Central Europe, within those regions which were ice-covered during the great glaciation, while the older, diploid forms have their habitat in the lower lying regions that were not formerly covered with ice. In *Empetrum* we have an example where the polyploid race, *E. hermaphroditum*, has a pronouncedly arctic distribution, while the diploid *E. nigrum* has a more southern habitat (HAGERUP, 1927). In this case the polyploid race has been made into a separate species. Several instances of this kind have been recorded by various authors (cf. MÜNTZING, 1936).

As known, the above-mentioned state of things, however, is not by any means an absolute rule, many cases in which conditions are the reverse having been met with. As extremes in this respect may be mentioned *Vaccinium uliginosum* (HAGERUP, 1933) and *Campanula rotundifolia* (BÖCHER, 1936). In these two species the diploids have a more marked arctic distribution than the polyploid races.

Apart from such intraspecific chromosomal races as those mentioned above, several authors have called attention to the fact that species

with high chromosome numbers seem to have a more northernly or alpine habitat than closely related species, within the same genus or family, with lower chromosome numbers. TÄCKHOLM (1922) first demonstrated this difference in geographical distribution between species with different chromosome numbers in the genus *Rosa*, species with the most northern area of distribution having the highest number, while species with lower numbers are more confined to a warmer climate. With regard to the *Bicornes* group HAGERUP (1928) was able to show that the species growing farthest to the north were invariably those with the highest polyploid numbers. Similar conditions seem to prevail within the *Cactaceae* (cf. STOCKWELL, 1935).

This interesting correlation between polyploidy and geographical distribution has been extensively studied by HAGERUP (1928, 1931) and especially by TISCHLER (1934). These investigators have emphasized the fact that polyploidy is a very important means, which in many cases renders possible an advance towards and an occupation of climatically unfavourable regions in which the diploids do not thrive. In other words the polyploids seem to be better adapted for extreme environmental conditions than the diploids. Consequently, the flora in such unfavourable regions should be richer in polyploids than usual, and TISCHLER has shown this to be the case in a comparison between the flora of Sicily and Iceland. While only 31 per cent of the examined species from Sicily were polyploids, the corresponding figure for Iceland was 55 per cent. Illustrative examples of this tendency to an increased number of polyploid elements in the flora from climatically unfavourable regions have been recorded recently by SOKOLOVSKAJA and STRELKOVA (1938). These authors have made extensive cytological studies of the flora of the high-alpine regions of Pamir and Altai. In Pamir, where very unfavourable conditions, climatic as well as all other growth requirements, prevail, polyploids were predominant, viz. 85 per cent polyploids among altogether 150 species examined. From the high-alpine zone of Altai, where the climate is somewhat milder than in Pamir, 200 species were examined, and of these 65 per cent were found to be polyploids.

Although an extensive material is already available to throw light on various problems in connexion with polyploidy, opinions still differ not the least as to the relation between polyploidy and geographical distribution. In connexion with the above-mentioned observations the present investigation of the high-arctic flora from Spitzbergen may therefore be of interest.

The material on which this investigation is based was collected at Isfjorden. The great majority of the chromosome numbers was determined in root-tip preparations. The fixative employed was LA COUR's 2 BD or other fixatives containing osmic acid. A few sections were fixed in »Navashin». The stain used was gentian violet.

Some of the chromosome numbers found in material from Spitzbergen have already been published, viz. those for the genus *Ranunculus* and the family *Gramineae* (FLOVIK, 1936, 1938), but in a complete survey like this it may be of interest to include also those in the list. The species examined and their chromosome numbers are tabulated in the following list — the diploids are denoted by an asterisk (*).

Species	2n
Gramineae.	
Alopecurus alpinus SM. ...... 112 + 3ff, 114 + 2ff, about	130 + 1f
Arctagrostis latifolia (R. BR.) GRISEB.	62
Arctophila fulva (TRIN.) RUPR.	42
Calamagrostis neglecta (EHRH.) P. B.	28
Deschampsia alpina ROEM. et SCHULT. 39, 41,	49
Dupontia Fisheri R. BR.	88 + ff
Dupontia Fisheri R. BR. var. *psilosantha* (RUPR.) SCHOL. ..	44 + ff
Festuca rubra L. var. *arenaria* (OSB.) E. FRIES	42 (+ 1f)
Festuca ovina L. var. *brevifolia* (R. BR.) HART	28
Festuca ovina L. var. *vivipara* L.	49
Hierochloe alpina (LILJEBL.) ROEM. et SCHULT.	56
Phippsia algida (SOLAND.) R. BR.	28
Phippsia concinna (TH. FRIES) LINDEB.	28
Phippsia concinna (TH. FRIES) LINDEB. var. *algidiformis* (SMITH) HOLM.	29
Poa abbreviata R. BR.	76 ±
Poa alpigena (E. FRIES) LINDM. 84,	77 ±
Poa alpigena (E. FRIES) LINDM. var. *colpodea* (TH. FRIES) SCHOL.	51 + 5ff
Poa alpigena (E. FRIES) LINDM. var. *vivipara* (MALMGR.) SCHOL.	42 + 4ff
Poa glauca VAHL.	70—72
Poa alpina L. var. *vivipara* L. 44,	42 + 4ff
Poa arctica R. BR.	56
Poa arctica R. BR. var. *vivipara* (MALMGR.) SCHOL.	56
Puccinellia angustata (R. BR.) RAND. et REDF.	42

Puccinellia phryganodes (TRIN.) SCRIBN. et MERR. 28
Puccinellia Vahliana (LIEBM.) SCRIBN. et MERR. 14*
Trisetum spicatum (L.) RICHT. 28

Betulaceae.

Betula nana L. 28

Polygonaceae.

Oxyria digyna (L.) HILL. 14*
Polygonum viviparum L. .... about 100

Caryophyllaceae.

Honckenya peploides (L.) EHRH. 66
Stellaria longipes GOLDIE 104
Stellaria humifusa ROTTB. 26*
Cerastium Regelii OSTENF. 72
Silene acaulis L. 24*

Ranunculaceae.

Ranunculus Pallasii SCHLECHT. 32
Ranunculus lapponicus L. 16*
Ranunculus Pallasii × *R. lapponicus* 24
Ranunculus hyperboreus ROTTB. 32
Ranunculus pygmaeus WAHLENB. 16*
Ranunculus nivalis L. 48
Ranunculus sulphureus SOLAND. 96

Papaveraceae.

Papaver radicatum ROTTB. 70

Cruciferae.

Cardamine pratensis L. 64
Draba alpina L. 80
Draba rupestris R. BR. 48
Draba arctica WAHLENB. 80
Cochlearia officinalis L. var. *groenlandica* (L.) GELERT. ... 14*

Saxifragaceae.

Saxifraga stellaris L. var. *comosa* RETZ. 56
Saxifraga nivalis L. 60
Saxifraga nivalis L. var. *tenuis* WAHLENB. 20*
Saxifraga hieraciifolia WALDST. et KIT. 112
Saxifraga oppositifolia L. 52

Saxifraga flagellaris WILLD.   32
Saxifraga hirculus L.   32
Saxifraga rivularis L. 26*
Saxifraga groenlandica L.   80
Chrysosplenium alternifolium L. var. *tetrandum* LUND. 24*

   Rosaceae.

Potentilla pulchella R. BR.   28
Potentilla emarginata PURSH.   42
Dryas octopetala L. 18*

   Empetraceae.

Empetrum hermaphroditum (LANGE) HAGERUP   52

   Ericaceae.

Vaccinium uliginosum L. 24*

   Polemoniaceae.

Polemonium boreale ADAMS 18*

   Campanulaceae.

Campanula rotundifolia L. 34*

   Compositae.

Erigeron uniflorus L. var. *unalaschkensis* (D. C.) OSTENF. . .   36
Petasites frigidus L.   60
Arnica alpina LAEST.   56
Taraxacum arcticum DAHLST.   40

Owing to the existing uncertainty as to the origin of the different chromosome numbers in the *Cyperaceae*, the numbers found in representatives of this family have not been included in the above list in so far as the recording of the polyploid elements is concerned. Of representatives of other families of higher plants the investigation comprises 68 species and varieties, 54 of which are polyploids and 14 diploids, i. e. about 80 per cent polyploids.

This high percentage of polyploids among the Spitzbergen flora agrees very well with the investigations of chromosome numbers in plants from other regions with extreme climatic or otherwise unfavourable conditions of growth, referred to above. The facts mentioned above may be interpreted as an evident tendency indicating that an increased chromosome number increases the adaptability to unfavourable habitats.

It is a well-known fact that an increase in the number of chromo-

somes frequently brings about considerable physiological and other changes in character (cf. MÜNTZING, 1936). — HAGERUP (1931, 1933) has pointed out in his works that with polyploidy there may follow a change in the ecological and plant-geographical value of the plant. Owing to its immediate revolution an increase in chromosome number may therefore be conceived to create at one stroke new forms that are more suited than the diploids to immigrate into and colonize new regions. But perhaps the greatest significance of an increase in the number of chromosomes lies in the fact that *it creates a foundation for a wider range of variation, which in turn leads to greater possibilities for the selection of types capable of colonizing regions where the adaptability of the diploids is inadequate to do so.* As a natural consequence of this increased range of variation the polyploids thus have as a rule a more extensive distribution than the diploids. But even to this rule there are exceptions. Owing to the gigantism that polyploidy frequently brings about, the possibilities of distribution may decrease. On the other hand, though many diploids are only of local importance, others are spread over large areas, partly with highly varying conditions of life, such as was shown by TURESSON (1938).

Among the examined species from Spitzbergen there are several that are worth while mentioning more in detail. This is especially true of the examples of intraspecific chromosome races met with. Of these, *Dupontia Fisheri* and *D. Fisheri* var. *psilosantha* have already been reported in detail (FLOVIK, 1938). Although both forms are typically arctic, the high chromosomal *D. Fisheri* seems to extend farthest to the north and otherwise occupies more unfavourable regions than var. *psilosantha*, from which it may have originated by a simple chromosome doubling.

In *Honckenya peploides* ROHWEDER (1936) found the chromosome numbers 2n = 48 and 64. In this species from Spitzbergen a divergent, and at the same time the highest, number, 2n = 66, was found. This aneuploid number does not seem to be a chance occurrence, for it was also established for certain in material of *H. peploides* from Tromsö (arctic Norway). The obviously constant occurrence of this aneuploid number is possibly due to apomixis, which appears to be quite common in arctic plants.

In material of *Cardamine pratensis* from Cambridge MANTON (1932) found two different chromosome numbers, viz. 2n = 32 and 64. In Spitzbergen only the 64-chromosome race was found, i. e. the one with the higher multiple.

The tetraploid *Empetrum hermaphroditum*, which is the sole prevailing form in East Greenland (HAGERUP, 1927), occurs also in Spitzbergen, while, as already mentioned, the diploid race, *E. nigrum*, has a more southern distribution.

Within the *Saxifragaceae* there are several examples of intraspecific chromosome races. *Saxifraga stellaris* was found by various authors to have the somatic chromosome number 28. This number was found by SKOVSTED (1934) in material. from Norway, by BÖCHER (1938) in material from Swedish Lapland, and by the present writer in material from Tromsö (arctic Norway). In *S. stellaris* var. *comosa* BÖCHER (l. c.) found 2n = 56 in material from Lapland, and this number was also found in the present material from Spitzbergen. When the chromosome numbers and the morphology of the chromosomes are taken into consideration, the most obvious conclusion is that var. *comosa* has arisen from *S. stellaris* by chromosome doubling. HARMSEN (1939) also assumes that var. *comosa* is a polyploid form of *S. stellaris* or a closely related species. The number 2n = 64, which was found by HARMSEN in the embryo of var. *comosa* from Greenland, is no evidence against this assumption (cf. HARMSEN, l. c.). The two forms differ with respect to ecology and geographical distribution. Var. *comosa*, having the higher number of chromosomes, is circumpolar, high-arctic (-alpine), whereas *stellaris* is subarctic — temperate — alpine.

In material of *Saxifraga nivalis* cultivated in the Botanical Garden of the University of Copenhagen SKOVSTED (1934) found the chromosome number 2n = 28, and in *S. nivalis* var. *tenuis* from Lapland (Sweden) BÖCHER (1938) found the number 2n = 20. In the examined material from Spitzbergen the numbers found are 2n = 60 and 20 for *nivalis* and var. *tenuis* respectively. With reference to the number 2n = 28 found by SKOVSTED, BÖCHER thinks it possible that in this case the basic number is 7 and that the number, 2n = 20, found in var. *tenuis* may have arisen by a cross between two *nivalis* races having the chromosome numbers n = 7 and 14 respectively. The numbers found in the Spitzbergen material, however, furnish stronger support for the assumption that the basic number is 10. — The external morphological characters, which refer essentially to quantitative differences — *nivalis* is more robust —, as well as the fact that no chromosome-morphological differences can be shown between the two forms may indicate that *nivalis* has arisen from var. *tenuis* by autopolyploidy. The polyploid form has a wider area of distribution than the diploid. This is especially true of the vertical distribution in that the polyploid

form goes farther down into the lowland as well as higher up on the mountains than the diploid form. Both forms occur in Spitzbergen, but according to SCHOLANDER (1934) *nivalis* is found somewhat farther north than var. *tenuis*. The two forms are also otherwise different with respect to ecology (cf. ARWIDSSON, 1938).

With regard to *Saxifraga hieraciifolia* from the high-alpine regions of Pamir and Altai, SOKOLOVSKAJA and STRELKOVA (1938) report the chromosome number 2n = 80—82. On the other hand, the same species from Spitzbergen was found to have 2n = 112 chromosomes. SOKOLOVSKAJA and STRELKOVA point out that in their investigations they occasionally came across polyploids with higher chromosome numbers in the arctic zone than in the mountainous regions. *S. hieraciifolia*, mentioned above, furnishes a new example of this. In *Saxifraga hirculus* from the same localities, Pamir and Altai, these authors found the chromosome number 2n = 28, while the chromosome number of this species from Spitzbergen was found to be 2n = 32.

In material of *Saxifraga oppositifolia* from Norway SKOVSTED (1934) found the number 2n = 26. The same species from Spitzbergen, on the other hand, was found to have 2n = 52 chromosomes. Still another example showing that the northernmost representatives have higher chromosome numbers than those having a more southern habitat.

On the question of the correlation between chromosome number and geographical distribution it has been repeatedly pointed out by various authors that it is not an absolute rule that representatives with high chromosome numbers are the most extreme arctic or alpine, and there are also interesting examples of this among the Spitzbergen flora. As already mentioned, we know the extreme instances of this kind as represented by *Vaccinium uliginosum* and *Campanula rotundifolia* from the investigations of material from Greenland and other places carried out by HAGERUP and BÖCHER. Both these species occur as diploids also in Spitzbergen, whereas representatives of the same species growing farther south are tetraploids. A new, analogous case is furnished by *Chrysosplenium alternifolium*. In discussing the typical arctic form, *C. alternifolium* var. *tetrandum*, SCHOLANDER (l. c., p. 18) writes as follows: »It would be of considerable interest to know the chromosome numbers of this little four-staminate form as compared with the southern larger and eight-staminate *C. alternifolium*. It would not be very surprising if the latter may be tetraploid as compared with the former». The cytological examinations now prove that SCHOLANDER's assumption was correct. The chromosome number in var. *tetrandum* from Spitz-

bergen was found to be 2n = 24, while the more southern *alternifolium*, according to SKOVSTED (l. c.), has the number 2n = 48.

Another new example of the same kind is provided by *Cochlearia officinalis*. In material of this species from the S. W. coast of Wales CRANE and GAIRDNER (1923) found the tetraploid number 2n = 28. In *C. officinalis* var. *alpina* from the Faroe Islands BÖCHER (1938) found the divergent number 2n = 26. On the other hand, in material of *C. officinalis* var. *groenlandica* from Spitzbergen, whose habitat is mainly confined to the northernmost polar regions, the diploid number 2n = 14 was found.

These last-mentioned findings need not, however, alter the main impression. The percentage of polyploids in this high-arctic flora is unusually high and, as already pointed out, should be interpreted as an evident tendency indicating that an increased chromosome number increases the adaptability to extreme environmental conditions. *Polyploidy must therefore have played an important role in the origin of the forms or species that have colonized the arctic regions.*

On the subject of polyploid intraspecific chromosome races MÜNTZING (1936) has rectified a lot of important data which affords a foundation for the conclusion that the majority of these races are autopolyploids, and that autopolyploidy or a purely quantitative increase in chromosome number has played a very important role in the evolution of higher plants. The above-mentioned intraspecific chromosome races should supply still further examples to MÜNTZING's already representative list of cases which strictly justifies this conclusion.

In modern floristic literature several of the chromosome races recorded here as varieties of the same species are separated as distinct species. The differences found in chromosome number furnish a new diagnostic character for this differentiation.

SUMMARY.

1. An investigation has been made of the chromosome numbers in higher plants from Spitzbergen. Altogether 68 species and varieties have been investigated. A list of these species and their chromosome numbers is given on pp. 432—434.

2. Of the species and varieties examined 14 are diploids and 54 polyploids, i. e. about 80 per cent polyploids. The high percentage of polyploids among this high-arctic flora agrees very well with the findings of other authors in their investigations of the chromosome

numbers of plants from other regions with extremely unfavourable climatic conditions. This may be interpreted as an evident tendency in the direction that an increased chromosome number increases the adaptability to extreme habitats, like arctic and alpine conditions. Polyploidy should therefore have played an important role in the origin of those forms or species which have colonized such regions as the arctic or alpine.

3. Several instances of intraspecific chromosome races, some of them new, were met with in the Spitzbergen flora. In most cases the high chromosomal race has the most northern range of distribution, but there are also examples showing the reverse of this. New instances of the latter are furnished by *Chrysosplenium alternifolium* and *Cochlearia officinalis*, both of which occur with diploid forms in Spitzbergen, while the more southern forms are polyploids. As has been pointed out by several other authors, this implies that there is no absolute rule that high chromosome number is associated with fitness to extreme habitats.

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EINE SERIE MULTIPLER ALLELE FÜR BLÜTENZEICHNUNGEN BEI GODETIA AMOENA

VON *GUNNAR HIORTH*

GENETISCHES INSTITUT DER LANDWIRTSCHAFTLICHEN HOCHSCHULE, AAS, NORWEGEN

I.

GODETIA ist eine beliebte einjährige Zierpflanze, die in zahlreichen Sorten kultiviert wird. Im Samenkatalog der Firma Benary, Erfurt, für das Jahr 1939 werden z. B. 42 Sorten verzeichnet. Abgesehen von der Sorte »Blauer Zwerg», die einer chilenischen Art entstammt, gehören alle diese Typen zu den beiden kalifornischen Arten *Godetia Whitneyi* und *Godetia amoena*. Die Gartenrassen dieser beiden Arten sind im Durchschnitt auffällig voneinander unterschieden:

Godetia Whitneyi.	*Godetia amoena.*
Niedrig — mittelhoch; buschig, stark verzweigt mit dichtem Blütenstand.	Hoch; lockerer, weniger verzweigter Habitus mit lockerem Blütenstand.
Kronblätter nie mit Fleck direkt an der Basis.	In der Regel mit Fleck direkt an der Basis.
Antheren nicht so satt gefärbt.	Antheren satt rot mit sattgelber Spitze.
Kapseln dick, fleischig, erst spät und nur bei günstigem Wetter trocknend, oft völlig geschlossen bleibend.	Kapseln dünn, leicht trocknend, bei Reife weit aufspringend.
	Samen kleiner.

Die Artzugehörigkeit der Gartensorten lässt sich in der Regel leicht bestimmen. Im Samenkatalog von Benary sind z. B. sämtliche 60 cm hohen oder höheren Sorten der Art *amoena*, sämtliche niedrigeren der Art *Whitneyi* zuzurechnen. Zweifel über die Artzugehörigkeit einer Sorte lassen sich übrigens leicht durch Kreuzung entscheiden, da der Bastard zwischen beiden Arten nahezu steril ist.

Die genannten Unterschiede sind jedoch nur bei der Einteilung der Gartenformen verwendbar. Über die Charakterisierung der natürlichen Formen von *Whitneyi* und *amoena* hat bis in letzter Zeit grosse Unsicherheit geherrscht. Kreuzungsversuche mit natürlichen Formen, die ich im Jahre 1936 eingesammelt habe, gestatten indessen, diese Arten genauer zu definieren. Über diese Untersuchungen wird eine besondere Abhandlung erfolgen. Einige Konklusionen seien indessen schon an dieser Stelle mitgeteilt.

Nach der neuesten systematischen Bearbeitung der Gattung *Godetia*
von HITCHCOCK (1930) kommt *Godetia amoena* an der Küste des Stillen
Ozeans von Monterey (150 km südlich von San Francisco) bis zur Süd-
spitze der Vancouver-Insel in British Columbia, Kanada, vor, während
für *G. Whitneyi* nur ein einziger Fundort, Shelter Cove (Humboldt Co),
an der Küste des nördlichen Kaliforniens angegeben wird. Dem gegen-
über ergaben meine Versuche folgendes:

1) Es besteht eine scharf umgrenzte Gruppe von *Godetia*-Arten,
die *amoena-Gruppe*, die mit keiner anderen Gruppe verwechselt werden
kann. Es erscheint zweckmässig, diese Gruppe in 3 Arten aufzuteilen,
G. nutans, *G. Whitneyi* und *G. amoena*. Erstgenannte Art ist durch
hängende Blütenknospen charakterisiert, während die beiden letzteren
aufrechte Knospen haben. Die Bastarde zwischen Formen mit auf-
rechten und mit hängenden Knospen sind stets nahezu steril.

2) Die Formen der *amoena*-Gruppe mit aufrechten Knospen sind
durch eine geographische Sterilitätsgrenze in zwei Arten geteilt, indem
die Formen südlich der Meeresbucht Golden Gate bei San Francisco mit
den Formen nördlich von Golden Gate sterile Bastarde geben. Wegen
der grossen Variabilität der nördlich von Golden Gate befindlichen
Art fällt es dagegen sehr schwer, diese Sterilitätsgrenze mit einwand-
freien morphologischen Unterschieden zu korrelieren. Indessen wurde
ein eigentümlicher Unterschied in der Blütenzeichnung gefunden. Ein
kleiner Fleck *direkt* an der Basis des Kronblattes (*Basalfleck*) ist die
vorherrschende Zeichnung südlich von Golden Gate. Ein mehr oder
weniger weit von der Basis entfernter Fleck (*Zentralfleck*) herrscht
nördlich von Golden Gate vor. Unter zahlreichen Populationen südlich
von Golden Gate habe ich nie eine Pflanze mit Zentralfleck angetroffen,
unter noch weit mehr Populationen nördlich von Golden Gate nie einen
Basalfleck. Individuen ohne jeglichen Fleck kommen dagegen in stark
wechselnder Häufigkeit im ganzen Verbreitungsgebiet der *amoena*-
Gruppe vor. — Auch in der Antherenfarbe scheint ein gewisser Unter-
schied zwischen beiden Arten zu bestehen, indem die Rassen südlich
von Golden Gate ein leuchtendes Rot mit satt gelber Spitze haben, die
Rassen nördlich davon weniger satte Farben.

3) Die kultivierten *Whitneyi*-Rassen geben fertile Bastarde mit den
Formen nördlich von Golden Gate, die kultivierten *amoena*-Rassen da-
gegen mit denen südlich von Golden Gate. Es scheint mir daher zweck-
mässig, die nördlich von Golden Gate vorkommende Art als *G. Whitneyi*,
die südlich davon vorkommende als *G. amoena* zu bezeichnen.

4) Die auffälligen Unterschiede zwischen kultivierten *Godetia*

amoena- und *G. Whitneyi*-Typen dürften zum grössten Teile auf Eigentümlichkeiten der Lokalrasse von *G. Whitneyi* beruhen, die zum Ausgangsmaterial für die Zierpflanzen dieser Art wurde, zum Teil auch auf Auslese bei der Züchtung. Die Zierformen von *amoena* haben in ihrem Habitus relativ grosse Ähnlichkeit mit den natürlichen Rassen dieser Art, während die kultivierten *Whitneyi*-Formen von den meisten wilden *Whitneyi*-Rassen stark abweichen.

Die Versuche mit *Godetia* wurden mit Handelssorten von *Whitneyi* und *amoena* im Jahre 1931 begonnen und 1936 durch Einsammlung wilder Rassen erweitert. Die Versuche mit *amoena* wurden in der Hauptsache auf die Analyse einer Serie multipler Allele für Blütenzeichnungen beschränkt und werden mit der vorliegenden Mitteilung im wesentlichen abgeschlossen.

Als Versuchsobjekte sind *G. Whitneyi* und *G. amoena* etwa gleich bequem. *G. Whitneyi* hat indessen den Vorteil einer weit grösseren Verbreitung in der Natur und einer vielfach grösseren Variabilität. Die Versuche werden nunmehr mit dieser Art allein fortgesetzt. — Es ist indessen wahrscheinlich, dass für genetische Versuche die extrem variable Gruppe *Godetia quadrivulnera-purpurea* noch besser geeignet wäre.

II.

Während ein Zentralfleck in natürlichen *amoena*-Populationen nie angetroffen wurde, kommt er bei einigen Handelssorten dieser Art vor. Dies liesse sich durch folgende Annahmen erklären: Der Zentralfleck könnte in anderen Teilen des Verbreitungsgebietes dieser Art vorkommen. Er könnte durch Kreuzung von den Arten *G. nutans* oder *G. Whitneyi* erworben sein oder innerhalb der Gartensorten durch Mutation entstanden sein. — Bei *G. nutans* wurden sowohl Rassen mit Zentralfleck als auch solche mit Basalfleck angetroffen, allerdings an weit voneinander entfernten Standorten.

Im Folgenden wird gezeigt, dass die Fleckformen der Gartenrassen von *amoena* durch eine Serie von multiplen Allelen bedingt werden. Es wurde mit folgenden Typen gearbeitet:

$f\ Ag$ = ohne Fleck, Antheren rot.

$F^{bsw}\ Ag$ = schwacher Basalfleck, Antheren rot.

$F^{bst}\ Ag$ = starker Basalfleck, Antheren rot.

$F^{k}\ ag$ = kleiner Zentralfleck, Antheren gelb.

$F^{m}\ ag$ = mittlerer Zentralfleck, Antheren gelb.

$F^m\,Ag$　＝ mittlerer Zentralfleck, Antheren rot.

$F^g\,ag$　＝ grosser Zentralfleck, Antheren gelb.

Ag-Pflanzen haben leuchtend rote Antheren mit grösserer oder kleinerer gelber Spitze, ag gelbe Antheren oder sehr schwach rote mit
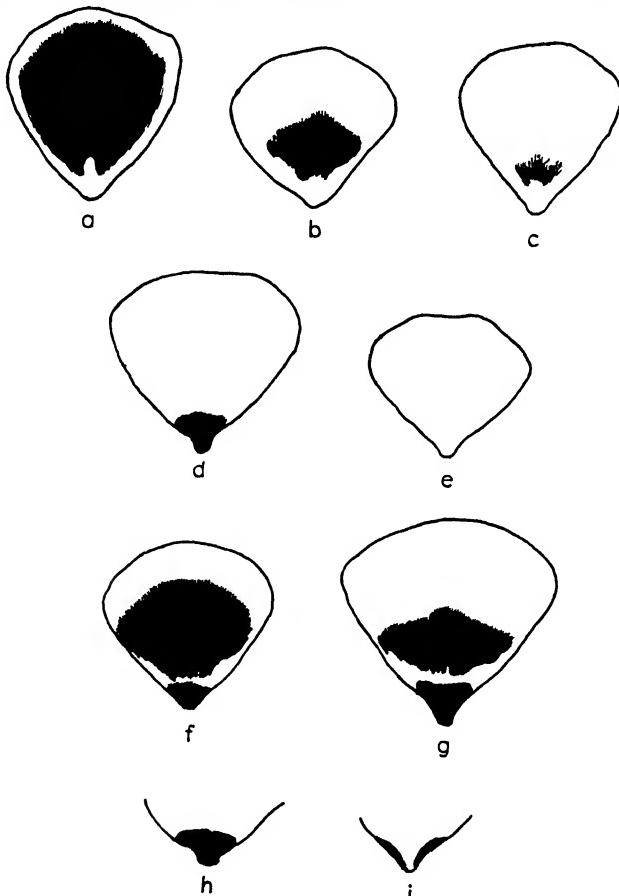

Fig. 1. Kronblattzeichnungen bei *Godetia amoena*. — *a*: Grosser Zentralfleck F^kF^k; *b*: mittlerer Zentralfleck F^mF^m; *c*: kleiner Zentralfleck F^kF^k; *d*: starker Basalfleck $F^{bst}F^{bst}$; *e*: ohne Fleck ff; *f*: F^gF^{bst}; *g*: F^mF^{bst}; *h* und *i*: Aussenseite (Unterseite) der Kronblattbasis von F^{bst} bzw. F^{bsw}.

gelber Spitze. Wenn auch die Färbung der »gelben Antheren» etwas variiert, ist der Unterschied zwischen Ag und ag auffällig und scharf.

f, das nicht aus Gartenmaterial sondern aus einer wilden Rasse stammt, bedingt vollständigen Fleckmangel.

F^{bsw}-Individuen besitzen einen relativ kleinen Fleck an der Basis des Kronblattes, F^{bst} einen etwas grösseren, stärker gefärbten (Fig. 1 *d*).

Die verschiedenen Typen von Zentralfleck sind bei *amoena* etwas näher an der Basis gelegen als bei *Whitneyi*. F^k-Individuen haben einen in Form und Grösse stark variablen Fleck. Dieser besteht aus kurzen roten Streifen, Strichen oder Punkten, die, wenn in geringer Anzahl vorhanden, nur ein kleines gestricheltes Areal bilden, wenn sie zahlreicher und dichter sind, zu einem fast kompakten Fleck zusammenfliessen, der nahezu die Grösse eines mittleren Zentralfleckes erreichen kann. Der durchschnittliche Unterschied zwischen F^k und F^m ist jedoch beträchtlich. Bei den übrigen Typen des Zentralfleckes sind nur die Ränder in Spitzen, Striche oder Punkte aufgelöst, die in der Richtung der Nervatur des Kronblattes verlaufen, welche von der Basis des Kronblattes fächerförmig zum distalen Rand ausstrahlt. Bei F^m-Pflanzen ist der Fleck ziemlich gross und kräftig gefärbt (Fig. 1 *b*). Bei F^g ist er noch grösser, sodass nur schmalere oder breitere Säume an den Rändern des Kronblattes freibleiben (Fig. 1 *a*).

Ausser obigen Allelen dürfte es noch eine Reihe anderer geben, die z. B. intermediäre Fleckgrössen zwischen den oben genannten bedingen.

Genauere Bestimmungen der Farben sind nicht vorgenommen worden. Die Grundfarbe (Farbe des ungefleckten Teils des Kronblattes) ist ein sehr helles Violett, während der Zentralfleck satt rotviolett gefärbt ist. Der Basalfleck hat in der Regel einen etwas abweichenden roten Farbton.

III.

Die Allelie der genannten Gene wurde durch die Kreuzungen in Tabelle 1 bewiesen. Zur Vereinfachung der Darstellung wurde im allgemeinen nicht zwischen $F^m Ag$ und $F^m ag$, ferner zwischen F^{bst} und F^{bsw} unterschieden. Die beiden letzteren Allele wurden dann unter der Bezeichnung F^b zusammengefasst. Es wurde jedoch mit allen diesen Typen gearbeitet und ein deutlicher Unterschied zwischen ihnen festgestellt.

Nr. 1—4 (Tabelle 1) zeigen die Spaltungszahlen in F_2 der Kreuzungen $F^g \times f$, $F^m \times f$, $F^k \times f$ und $F^b \times f$. Wir erhalten stets rund ein Viertel ff-Individuen. f ist jedoch nicht völlig rezessiv, da es in Heterozygoten die Grösse des Fleckes in der Regel etwas verkleinert und den Farbton etwas abschwächt.

F^g dominiert über F^m und lässt sich in F_2 sicher von ihm trennen (Nr. 5). Nr. 10 zeigt das Ergebnis der Rückkreuzung $F^gF^m \times ff$. Die beiden Typen in der Nachkommenschaft, F^gf und F^mf, waren in ihrer

Fleckgrösse so stark verschieden und variierten so wenig, dass jedes
Individuum ohne Mühe identifiziert werden konnte. Dieser Befund
wurde durch die gleichzeitige Spaltung eines gekoppelten Gens bestätigt;
siehe unter Nr. 21.

Nr. 6 enthält eine Kreuzung $F^{bst} \times F^{bsw}$. Starker Basalfleck do-
miniert über schwachen und in F_2 liess sich jede Pflanze bequem aus-
zählen, was besonders dadurch ermöglicht wird, dass diese beiden
Allele eine verschiedene Zeichnung auf der Aussenseite der Kronblätter
bedingen (Fig. 1 *h, i*).

Kreuzungen von F^g, F^m und F^k mit F^b ergeben F_1-Pflanzen, die
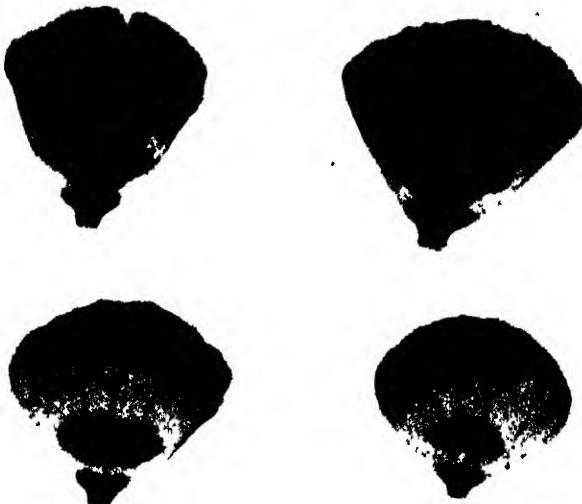
Fig. 2. Kronblätter von $F^m F^b$-Pflanzen aus F_2 von $F^m \times I^{.b}$. Lichtbild von getrock-
neten Kronblättern.

beiderlei Flecktypen nebeneinander zeigen. Wurde hierzu F^{bsw} be-
nutzt, so war der Basalfleck meist abgeschwächt und konnte in einigen
Blüten fast verschwunden sein. Wurde dagegen F^{bst} verwendet, so
zeigten sowohl $F^g F^b$-, $F^m F^b$- wie $F^k F^b$-Pflanzen einen sehr deutlichen
Basalfleck (Fig. 1 *f, g*). In F_2 (Nr. 7—9) ergab sich eine Spaltung nach
dem Schema

$$1\ F^x : 2\ F^x F^b : 1\ F^b$$

wobei F^x den betreffenden Typus von Zentralfleck bezeichnet (Fig. 3).

Eine Nr. 8 entsprechende Kreuzung ist schon von Rasmuson (1921)
mit demselben Ergebnis untersucht worden. Da ihm aber innerhalb

der Art *amoena* keine anderen Flecktypen zur Verfügung standen, speziell keine flecklose Rasse, konnte er den Nachweis der Allelie von F^m und F^b nicht vollständig erbringen.

Unter Nr. 11—13 finden wir die Ruckkreuzungen der Typen $F^g F^b$, $F^m F^b$ und $F^k F^b$ mit ff. Etwa die Hälfte der Nachkommen hat in jedem Fall den betreffenden Typus des Zentralfleckes, die andere Hälfte einen
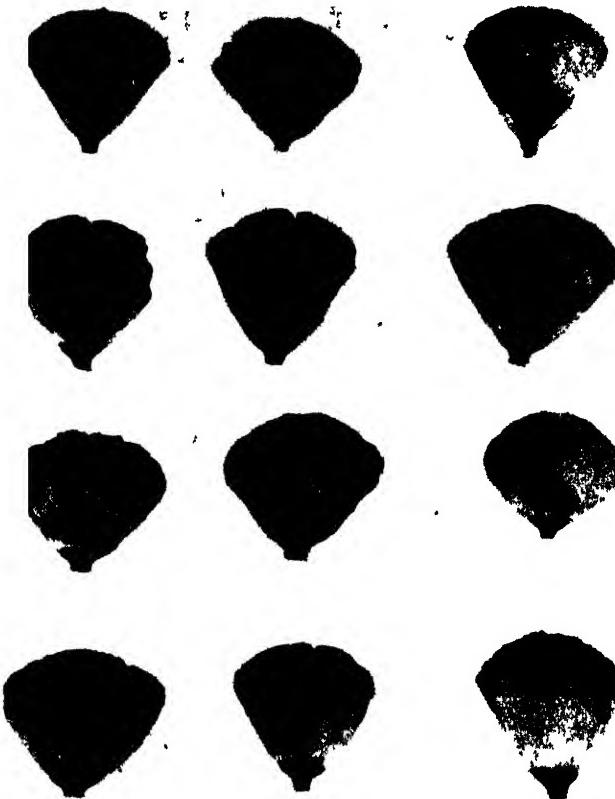
Fig 3   Kronblatter der Typen $F^m F^m$, $F^m F^b$ und $F^b F^b$ aus F_2 von $F^m \times F^b$.

Basalfleck. — Wollte man annehmen, dass Basalfleck und Zentralfleck nicht auf allelen sondern auf gekoppelten Genen beruhten, so musste hier eine besonders starke Koppelung vorliegen. Denn bei den Ruckkreuzungen Nr. 11—13 würde ein crossing-over sich durch das Auftreten von $F^x F^b$- und ff-Pflanzen verraten. Unter 3070 Nachkommen wurden keine derartigen Pflanzen gefunden.

Vereinzelte Ausnahme-Individuen mit Zentralfleck $+$ Basalfleck,

TABELLE 1. *Übersicht über die Spaltungen der Blütenzeichnungen bei Godetia amoena. Beschreibung der Merkmale siehe Abschnitt II, S. 443—445 *.*

	Kreuzung	F^x	f			n
Nr. 1.	$(F^g f)^2$	159	55			214
» 2.	$(F^m f)^2$	375	89			464
» 3.	$(F^k f)^2$	123	40			163
» 4.	$(F^b f)^2$	608	168			776
		F^k	F^m			
» 5.	$(F^g F^m)^2$	137	32			169
		F^{bst}	F^{bsw}			
» 6.	$(F^{bst} F^{bsw})^2$	106	31			137
		F^x	$F^x F^b$	F^b		
» 7.	$(F^g F^b)^2$	97	207	102		406
» 8.	$(F^m F^b)^2$	223	513	208		944
9.	$(F^k F^b)^2$	75	221	93		389
	Σ 7—9	395	941	403		1739
		F^g	F^m			
Nr. 10.	$F^g F^m \times ff$	186	190			376
		$F^\wedge$	F^b			
11.	$F^g F^b \times ff$	605	551			1156
12.	$F^m F^b \times ff$	568	532			1100
13.	$F^k F^b \times ff$	435	379			814
	Σ 11—13	1608	1462			3070
		$F^x ag$	$F^x F^b Ag$	$F^b Ag$		
Nr. 14.	$(F^g ag/F^b Ag)^2$	35	80	26		141
» 15.	$(F^m ag/F^b Ag)^2$	56	124	49		229
	Σ 14—15	91	204	75		370
		$F^g ag$	$F^g Ag$	$f Ag$		
Nr. 16.	$(F^g ag/f Ag)^2$	44	115	55		214
		$F^m ag$	$F^m F^b Ag$			
» 17.	$F^m ag/F^b Ag \times F^m ag F^m ag$ ..	178	216			394
		$F^x Ro$	$F^x ro$	$F^y Ro$	$F^y ro$	
Nr. 18.	$(F^x Ro/F^y ro)^2$ **	334	53	46	82	515
» 19.	$(F^x ro/F^y Ro)^2$	393	190	139	5	727
» 20.	$F^g ro/F^b Ro \times ff\, ro\, ro$	76	230	231	68	605
» 21.	$F^g ro/F^m Ro \times ff\, ro\, ro$	42	144	146	44	376
	Σ 20—21	118	374	377	112	981

die in zwei anderen Rückkreuzungsfamilien von diesem Typus auftraten, besassen, wie eine Nachprüfung zeigte, die Konstitution F^x/F^b und konnten demnach nicht durch crossing-over entstanden sein, sondern dürften auf Versuchsfehler beruhen.

Eine starke Koppelung der Antherenfarbe mit bestimmten Fleck-Allelen wurde in zahlreichen Kreuzungen konstatiert. Nr. 14—17 gibt das relativ geringe Material wieder, in dem die Antherenfarbe genau ausgezählt wurde. In Nr. 17 würde jeder Fall von crossing-over sich durch Entstehung von $F^m Ag$- oder $F^m F^b ag$-Pflanzen verraten, während in Nr. 14—15 nur die Hälfte der Crossovers entdeckt werden könnten. Zusammen befand sich daher unter ca. 580 Gameten kein Crossover-Gamet.

Da Ag ungefähr dieselbe rote Farbe auf den Antheren bedingt, wie F^g oder F^m auf den Kronblättern, dürfte es immerhin wahrscheinlicher sein, dass Antheren- und Kronblattfarbe von ein und demselben Gen bedingt werden als von zwei gekoppelten. Es liessen sich demnach auf Grund der Antherenfarbe einige weitere Allele für den Lokus F aufstellen; z. B. wären $F^m ag$ und $F^m Ag$ zwei F-Allele. — Übrigens bestärken die Befunde über die Antherenfarbe zugleich auch die Annahme von Allelie zwischen Basalfleck und Zentralfleck. Denn je mehr Gene mit Wirkung auf dasselbe Merkmal (lokalisierte rote Farbe) an derselben Stelle der Koppelungsgruppe gefunden werden, um so grösser ist die Wahrscheinlichkeit dafür, dass die gemeinsame Lage der Gene durch Allelie bedingt wird.

In zahlreichen Kreuzungen wurde schliesslich eine partielle Koppelung konstatiert zwischen den F-Allelen und einem Gen ro (rosa), das die hell violette Grundfarbe der Blüte in weisslich-rosa verwandelt. In Nr. 18 und 19 bezeichnen F^x und F^y verschiedene Fleck-Allele. Das Rekombinationsprozent (in Nr. 20—21) beträgt $230 : 981 = 23$ %.

Was die Konstanz der benutzten Fleck-Allele betrifft, so machen F^g, F^m, F^b und f durchaus den Eindruck von stabilen Genen. In den F_2-Familien von Nr. 1—9 und den Rückkreuzungen Nr. 10—13 traten keine Individuen auf, die den normalen Variationsbereich ihres Allels

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* F^x bezeichnet den in der Spalte Kreuzung an erster Stelle, F^y den an zweiter Stelle genannten Zeichnungstypus. F^b bezeichnet in einigen Fällen F^{bst}, in anderen F^{bsw}.

** Nr. 18 und 19 fassen eine Reihe von Kreuzungen zusammen, in denen verschiedene, hier nicht angegebene Allele spalten. Zur Vereinfachung der Darstellung wird hier F^y als rezessiv behandelt.

überschritten und etwa eine für ein anderes Allel typische Fleckgrösse zeigten. Indessen haben F^k und ein bestimmter, nicht näher daraufhin untersuchter Typus von F^m eine recht variable Fleckgrösse. Für F^k wurden nicht unbeträchtliche Unterschiede festgestellt zwischen verschiedenen Blüten einer Pflanze, verschiedenen Pflanzen einer Familie und zwischen verschiedenen Familien. Der Fleck kann winzig klein sein oder nahezu die Grösse eines mittleren Zentralfleckes erreichen. Der durchschnittliche Unterschied zwischen F^m und F^k ist jedoch sowohl bei Selbstungen als auch in F_2 bei Kreuzungen mit den gleichen Linien recht auffällig. Es liegt nahe anzunehmen, dass F^k ein labiles Gen ist, das innerhalb bestimmter Grenzen variiert.

Eine detaillierte Besprechung der Zahlenverhältnisse hat wenig Interesse, da bei *Godetia* statistisch bedeutsame Abweichungen nicht selten sind. Es dürfte genügen, auf einen Befund hinzuweisen. In Σ 7—9 treten die Heterozygoten in einer Häufigkeit von 54,1 % statt 50 $\pm$ 1,2 % auf. Abweichung: mittlerer Fehler = 3,4. Auch in Nr. 14—16 scheint die Häufigkeit der Heterozygoten erhöht zu sein. Da eine Begünstigung bestimmter Gametentypen stets zu weniger als 50 % Heterozygoten führen würde, muss hier nach einer anderen Erklärung gesucht werden. Man könnte annehmen, dass die Homozygoten mit zwei F-Chromosomen gleichen Ursprunges eine etwas stärkere Inzuchtswirkung zeigen und daher eine höhere Sterblichkeit haben als die Heterozygoten mit zwei F-Chromosomen verschiedenen Ursprunges.

IV. DISKUSSION.

Zwischen den Gliedern der Serie F^g, F^m, F^k, F^{bst}, F^{bsw}, f bestehen teils quantitative, teils qualitative Unterschiede. F^g, F^m und F^k bedingen verschiedene Grössen des Zentralfleckes; ein Zentralfleck ist aber durch seine Lage qualitativ von einem Basalfleck unterschieden.

Bei Kreuzungen quantitativ verschiedener Zeichnungen erhalten wir die üblichen Dominanzverhältnisse, der grössere Fleck ist (nahezu) dominant über den kleineren. Bei Verbindung qualitativ verschiedener Zeichnungen dagegen gibt es keine Dominanz, sondern die Heterozygoten zeigen beiderlei Flecktypen deutlich nebeneinander. Im Prinzip dürfte die Sachlage die gleiche sein, wenn die Kombination $F^m ag\ f\ Ag$ neben dem Zentralfleck von F^m die roten Antheren von f zeigt. In diesen Fällen wirken also die betreffenden Allele unabhängig voneinander, genau so wie es von mir (1931) bei qualitativ verschiedenen Blatt- und Kotyledonenzeichnungen bei *Collinsia* gefunden wurde.

Man könnte theoretisch erwarten, beim Studium von in der Natur vorkommenden Allelenserien relativ oft auf eine »unabhängige Wirkung» von Allelen zu stossen, indem die verschiedenen Allele einer bestimmten Serie durch eine Anzahl von Mutationsschritten, die im Laufe der Evolution des Gens nach und nach im selben Lokus stattgefunden haben, sich in stark divergierende Richtung entwickelt haben könnten (vgl. HIORTH, 1931, S. 262—266). Dies scheint auch der Fall zu sein. Verf. ist in drei Fällen auf eine unabhängige Wirkung von Allelen gestossen, nämlich bei Blatt- und Kotyledonenzeichnungen von *Collinsia*, bei den hier besprochenen Blütenzeichnungen von *Godetia amoena* und bei noch näher zu untersuchenden Kotyledonenzeichnungen von *Godetia Whitneyi*.

Inwiefern ein Zentralfleck innerhalb der Art *G. amoena* als natürlicher Charakter aufgefasst werden darf, bleibt allerdings nach den Angaben auf S. 443 fraglich. Daher sind in diesem Zusammenhang die Verhältnisse bei der verwandten Art *G. nutans* von Interesse, bei der sowohl Standorte mit Basalfleck, mit Zentralfleck und ohne Fleck vorkommen. Fleck gekreuzt mit ohne Fleck gibt 3 : 1-Spaltungen. Die Kreuzung einer Form mit Basalfleck (Magalia, Butte Co, Calif.) mit einer mit Zentralfleck (Glen Ellen, Sonoma Co, Calif.) ergab in F_2 eine Spaltung: 16 mit Zentralfleck : 46 Zentralfleck + Basalfleck : 21 mit Basalfleck, also ein Verhältnis 1 : 2 : 1. Demnach sind anscheinend auch bei dieser Art die Gene für Basalfleck und Zentralfleck allel und zeigen unabhängige Wirkung.

Dagegen sind nur wenig entsprechende Fälle in der Literatur erwähnt. Dies dürfte aber wohl darauf beruhen, dass für die meisten anderen Charaktere als lokalisierte Zeichnungen eine unabhängige Wirkung von Allelen nicht ohne besondere Untersuchungen nachgewiesen werden kann. Die »intermediäre Vererbung», die so allgemein bei Kreuzungen natürlicher Rassen angetroffen wird, liesse sich vielleicht bei genauester Betrachtung zum grossen Teil auf eine unabhängige Wirkung von Allelen zurückführen.

EAST (1936, S. 392) fand bei seinen Untersuchungen über Artbastarde bei *Nicotiana* weder Anzeichen dominanter noch rezessiver Gene, sondern es schien, dass »each gene is an active pattern former».

EAST unterscheidet zwischen defectiven und non-defectiven Allelen und führt die Heterosiserscheinungen hauptsächlich auf letztere zurück. Während in der Regel zwei *identische* non-defective Allele *AA* bei Homozygoten die gleiche Wirkung hätten wie ein einziges bei Heterozygoten *Aa*, könnten zwei *verschiedene* non-defective Allele (z. B. A_1A_2 oder A_3A_4) eine kumulative Wirkung entfalten. »The cumulative action of two non-defective allelomorphs of a given gene approaches the strictly additive as they diverge from each other in function.»

Es wäre interessant das *Godetia amoena*-Material im Sinne dieser Theorie auszuwerten. Dieses Versuchsobjekt scheint beträchtliche Vorteile für derartige Studien zu haben, insbesondere den, dass in den meisten benutzten Linien keine modifizierenden Gene vorkamen, die die Wirkung der Fleck-Allele in stärkerem Grade beeinflussen. Es war jedoch keine Gelegenheit, das Material eingehend genug zu studieren. Ein interessanter Befund liess sich indessen auch ohne Spezialuntersuchungen feststellen. Die Heterozygoten $F^k F^{bst}$ haben durchschnittlich einen weit stärkeren Zentralfleck als $F^k F^k$-Pflanzen. Hier liegt anscheinend nicht nur eine additive Wirkung zweier Allele vor, sondern auch eine darüber hinaus gehende Verstärkung.

V. ZUSAMMENFASSUNG.

1) Der Artname *Godetia amoena* wird hier auf die Rassen südlich von Golden Gate (San Francisco) mit aufrechten Knospen beschränkt.

2) Bei Gartenrassen von *Godetia amoena* wurde eine Serie multipler Allele für Blütenzeichnungen festgestellt. F^g, F^m, F^k bedingen einen grossen bzw. mittleren, kleinen »Zentralfleck«, F^{bst}, F^{bsw} einen starken bzw. schwachen »Basalfleck«, f vollständigen Mangel des Fleckes. Siehe Fig. 1 *a—e*.

3) Die fünf ersteren Allele sind unvollständig dominant über f; F^g ist (nahezu) dominant über F^m.

4) Die Kombinationen von F^g, F^m und F^k mit F^{bst} und F^{bsw} zeigen beiderlei Zeichnungstypen nebeneinander. Siehe Fig. 1 *f, g*; Fig. 2. *Bei Kreuzungen qualitativ verschiedener Zeichnungen wird also keine Dominanz, sondern eine »unabhängige Wirkung» der Allele angetroffen.* — F^{bst} vergrössert den Zentralfleck von F^k.

5) Rote (statt gelbe) Antherenfarbe ist stark mit bestimmten Fleck-Allelen gekoppelt. Unter ca. 580 Gameten wurde kein Fall von crossing-over angetroffen. Falls, wie es wahrscheinlich ist, rote Antherenfarbe und rote Blütenzeichnungen durch dasselbe Gen bedingt werden, muss die Anzahl der F-Allele erhöht werden.

6) Die F-Allele sind mit einem Gen für rosa-weisse Grundfarbe der Blüten gekoppelt mit einem Rekombinationsprozent von 23.

Fond, Norges Landbrukshöiskoles Forskningsfond) ermöglicht worden. Hierfür sei auch an dieser Stelle mein bester Dank ausgesprochen.

Aas, Norwegen, 20. Januar 1940.

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MEIOSIS OF ALLIUM PORRUM, A TETRA-PLOID SPECIES WITH CHIASMA LOCAL-ISATION

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WHILE meiosis of diploid forms with localised chiasmata has been analysed on several occasions within various Liliaceous genera (*Fritillaria, Allium, Trillium*), this characteristic course of meiosis has not been studied so far in any polyploid form. Such a study affords an approach to problems of very great interest. DARLINGTON (1937) points to one special problem: »In polyploid forms with localised pairing and chiasmata we must expect fewer multivalents than in corresponding forms with complete pairing, since the effective length for pairing is shorter» (l. c. p. 129). This conclusion is drawn from a study of diploid *Fritillaria* species, where the pachytene pairing in forms with local-isation of chiasmata only reaches about 50 % of the total length of the chromosomes. In *Allium*, on the other hand, the pachytene pairing is morphologically complete in localised species such as *Allium fistulosum* (LEVAN, 1933), it is complete even in asynaptic species, where no single chiasma is formed. Thus, in an autopolyploid *Allium* with localised chiasmata a normal frequency of multivalents may be expected at earlier meiotic stages, and this frequency should decrease during the course of the prophase. At metaphase I only such multivalents as are associated by chiasmata should be left, even if the mutual orientation of some bivalents should indicate that earlier they had been joined to quadrivalents.

During my work on the cytology of *Allium* I came across an auto-tetraploid species, *Allium Porrum* L., which showed almost complete localisation of chiasmata at meiosis. I studied several forms of the species, both Swedish commercial varieties and material procured from botanical gardens. All these forms were tetraploids and had localised chiasmata. In one form I found a regular occurrence of two small somatic chromosome fragments, which sometimes paired at meiosis, in other respects no cytological differences were found between the different forms.

In *Allium Porrum* ordinary NAVASHIN fixations of whole buds, preceded by a short dipping in CARNOY, gave better results than smears in osmic acid fixations. The pachytene stage in particular is very beautiful in NAVASHIN fixed material, permitting a detailed study of whole quadrivalents at this stage.

The somatic chromosomes of *Allium Porrum* are of the ordinary *Allium* type, 28 chromosomes have a median centric constriction and 4 chromosomes have a subterminal constriction. These latter are the s_1 chromosomes and their satellite is quite small. The length of the longest chromosomes of *Allium Porrum* is about 10 μ.

I. THE COURSE OF MEIOSIS.

As already mentioned, the prophase stages show complete chromosome pairing. Small unpaired segments may, of course, be found even at mid-pachytene, but they do not occur more frequently towards the chromosome ends. Such unpaired segments are found also in species with random-distribution of chiasmata. The chromomeric structure is very clear at pachytene, and the chromomeres are sometimes rather broad, almost band-like. There is one nucleolus in each cell. At the nucleolus two deeply stained lumps, evidently the end portions of the s_1 bivalents, may be seen quite regularly. No differences in the pairing can be seen between the proximal and the distal parts of the chromosomes. The diplotene loops, however, seem to appear at first in the distal parts of the chromosomes, and this might be an indication of a less intimate pairing in these parts.

During pachytene quadrivalents are easily found in most cells (Fig. 1 *a—b*). The exchange of threads within the quadrivalents usually occurs in one place, but also quadrivalents with two exchanges have been met with. If there is one exchange it may be located in any region of the chromosomes, medially or terminally. It is difficult at this stage to analyse whole cells, but I have often observed more than one quadrivalent in the same cell. Sometimes all the chromosomes seem to be joined into 8 quadrivalents.

At diplotene the quadrivalents are still present in great number (Fig. 1 *c—d*). The spiralisation is prominent and the quadrivalents are held together by the torsion. During diakinesis, however, many of the earlier quadrivalents are seen to fall apart into pairs of bivalents (Fig. 1 *e—h*). Now the remarkably regular distribution of chiasmata is clearly seen: always 2 chiasmata per bivalent, one at each side of

the centromere. Owing to the size of the chromosomes and their great
number diakinesis is not a suitable stage for studying the quadrivalents.

At the first metaphase the chromosomes are arranged regularly
into an equatorial plate. The characteristic structure of the bivalents
is now clear. Owing to the chromosome contraction, the two chiasmata
of each bivalent have been pulled together close to the centromere,
so the chromosomes seem to touch each other only at one point. In
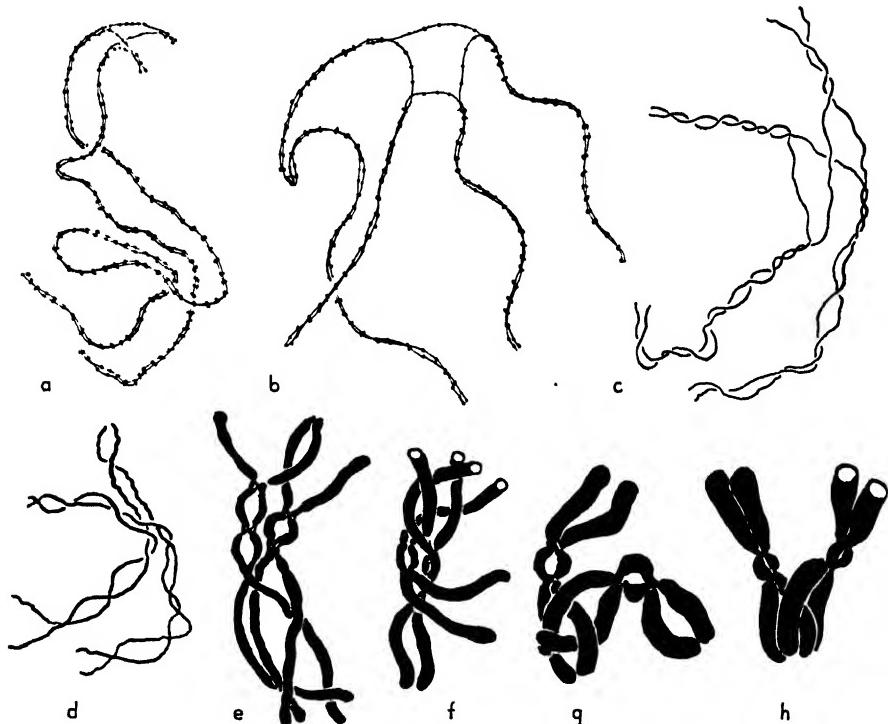
Fig. 1. a—b: quadrivalents at pachytene, c, d: at diplotene, e—h: quadrivalents at
diakinesis, held together only by torsion. — ✕ 3900.

polar view (Fig. 2 a) it is difficult to see the chiasmata but in side-view
(Fig. 2 b) the chromatid arrangement is made quite clear. Most plates
consist of only this kind of bivalents. The regularity is striking and
gives a good illustration of DARLINGTON's term »semi-clonal» in-
heritance.

There may occur, however, certain exceptions to the scheme. The
most common exception is the occurrence of bivalents with only one
chiasma (Fig. 2 c—d). In one pollen sac the following frequency of
such bivalents was counted:

Number of deviating bivalents:	0	1	2	3	4	5	Total	M/cell
Number of cases:	7	6	9	3	5	1	31	1,9

Compared with the conditions in *Allium fistulosum*, a diploid species with chiasma localisation, the frequency of deviating bivalents is rather high. *Allium fistulosum* had from 0,15 to 0,55 such bivalents per cell. In the next chapter it is suggested that the formation of pachytene
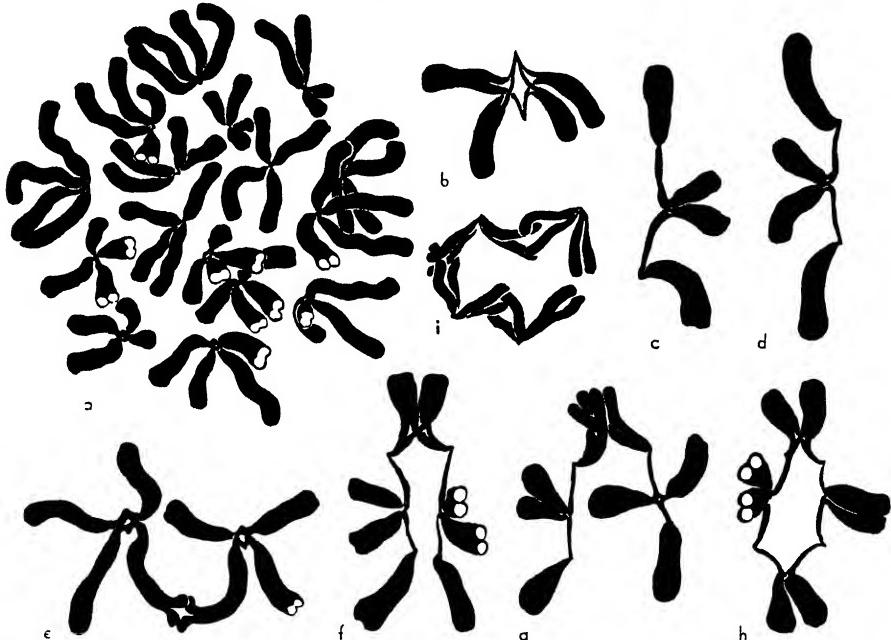
Fig. 2. *a*: metaphase I, a plate consisting of 16 cruciform bivalents seen from the pole, *b*: side-view of a cruciform bivalent, *c*, *d*: bivalents with one chiasma, *e*: a quadrivalent held together by a non-localised chiasma, *f*, *g*: localised chain quadrivalents, *h*: a localised ring quadrivalent, *i*: a ring quadrivalent at anaphase I. — *a*: × 1400, *b—i*: × 2800.

quadrivalents may predispose to the origin of pairs of bivalents with one chiasma.

In these bivalents with only one chiasma, usually the portion between the centromeres and the chiasma is extended and is somewhat narrower than the rest of the chromosomes. The chiasma is often more terminalised than in the cruciform bivalents within the same plate. The chromatid pairing evidently yields more easily under the strain from the centromeres in the bivalents with only one chiasma. In the cruci-·

form bivalents, where four chromatids resist the strain of the centro-
meres, the chiasmata seem to be more stationary.

Quadrivalents are present so rarely at metaphase I that an intense
study was needed merely to demonstrate their occurrence. This study
was made difficult by the large size of the 64 long cross-arms present,
which quite fill up the equatorial plates, so that the exact analysis of
each element is often rendered impossible. It was detected, however, that
now and then two cross-arms joined by a clear-cut chiasma were
stretched out extremely far towards one pole. In the cases where the
connections of these arms could be followed also inside the plate they
were found to form each one chiasma with two other chromosomes.
In fact there were present instances of chain quadrivalents of a type
hitherto unrecorded (Fig. 2 *f—g*). The chiasmata of these quadri-
valents were gathered as close to the four centromeres as possible.
After an extensive search the corresponding ring-type of quadrivalents
was also found (Fig. 2 *h*). These rings were observed only three times.
I wish to point out, however, that the occurrence of quadrivalents is
probably somewhat more frequent than these data indicate, since they
can be demonstrated only in especially favourable cases. Even if this
is taken into account the quadrivalents must be very rare at meta-
phase I, and certainly much rarer than in autotetraploids with random-
distribution of chiasmata. I counted in one slide 6 quadrivalents in 250
analysed cells and in another slide 4 quadrivalents in 130 cells. In
normal tetraploids of *Allium* the average frequency is 1—5 quadri-
valents per cell, i. e. about 100 times greater frequency.

The appearance of the quadrivalents is characteristic. They differ
from ordinary quadrivalents in the same qualities as localised bivalents
differ from ordinary bivalents. Thus the chiasmata of the quadri-
valents are very close to the centromeres, which brings about the form-
ation of large cross-arms at each chiasma. The orientation on the
spindle of the quadrivalents is typical, the 4 centromeres form a
rectangle with 2 corners at each side of the equator. Neighbouring
centromeres are orientated towards the same pole. Anaphase form-
ations indicate that sometimes also zig-zag arrangement may occur, but
such quadrivalents were not observed at metaphase. Anaphase of a
ring quadrivalent is seen in Fig. 2 *i*.

It was often noticed that in the quadrivalents the portions between
the centromeres and the chiasmata were longer and more extended
than the corresponding portion of the cruciform bivalents. The chi-
asmata of the quadrivalents were evidently more terminalised. This is

probably due to the same condition as that causing the greater terminalisation of the bivalents with one chiasma. It may be that also quadrivalents with their centromeres more close together really occurred, they would anyhow be very difficult to analyse. Formations which might be such quadrivalents were sometimes noticed, but they were interpreted as interlockings of two bivalents.

In the same manner as in *Allium fistulosum*, the localisation of the chiasmata was not absolute, non-localised chiasmata were observed also in *Allium Porrum*, although very seldom. Such chiasmata could give rise to quadrivalents of a new type. They were built up of two bivalents in which two cross-arms, one from each bivalent, were joined to a subterminal chiasma (Fig. 2 *e*). This chiasma did not influence the orientation of the quadrivalent on the spindle. The frequency of non-localised chiasmata was lower than in *Allium fistulosum*, where some forms had up to 1,7 non-localised chiasmata per cell. In one slide of *Allium Porrum* 3 such chiasmata were counted in 250 cells, and their average occurrence probably does not exceed 1 % of the cells.

Anaphase I and the second division take place normally. The pollen grains contain 16 chromosomes, so the few quadrivalents of meiosis evidently give rise to but slight disturbances.

II. DISCUSSION.

In the present paper a record is given of meiosis of an autotetraploid *Allium* species with almost absolute localisation of the chiasmata. DARLINGTON (1939) regards this proximal type of localisation only as a special case. Its type depends on the location of the original segment of pairing at zygotene. In other cases, for instance, tetraploid species of *Tradescantia*, chiasmata are formed only at the chromosome ends, due to the fact that the pairing starts distally.

Another independent genetic variable determining the meiosis type is the time limit of effective pairing. The time limit determines the degree of localisation. If the time of effective pairing is unlimited, the chromosomes will be paired along their whole length and their chiasmata will be distributed at random. The reason why the ends of the chromosomes of the proximal type of localisation do not form any chiasmata is, still according to DARLINGTON, not due to a precocious reproduction of the chromosomes in these parts, as HUSKINS and SMITH (1934) suggested, but is instead due to the time limit, which cuts off the effective pairing before it has reached the whole chromosome length..

The shorter this time limit, the more absolute is the localisation of chiasmata. In such asynaptic forms as *Allium amplectens* (LEVAN, 1940) the time limit must be nil. Since, however, a clear morphological pachytene pairing is seen both in localised and asynaptic *Allium* species,
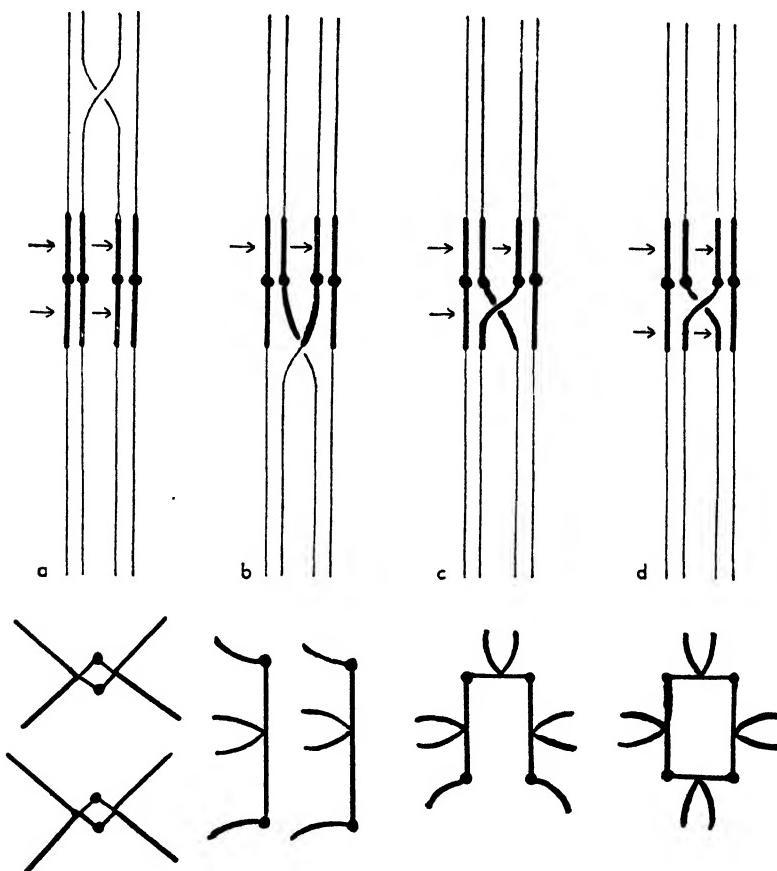
Fig. 3. Scheme of the possible cases of partner exchange in the pachytene quadrivalents and the result at metaphase I. Chiasmata are formed at the arrows. For further explanation see the text.

a distinction must be made between the apparent, visible pairing and the effective pairing, i. e. the pairing which gives rise to chiasmata.

It is clear that the original segment of pairing in such a species as *Allium Porrum* cannot be localised to a single exact point of the four homologous chromosomes. In that case quadrivalents could hardly be formed. The frequent occurrence of quadrivalents at pachytene indicates, in my opinion, that the pairing starts in different places even in

chromosomes with absolute localisation of chiasmata. A quadrivalent can be formed only if the pairing starts in at least two places within different pairs of the 4 homologous chromosomes. These starting points are possibly located close to the centromeres in *Allium Porrum,* otherwise the conclusion must be drawn that the zone of effective pairing is not always the zone of the earliest pairing.

However this may be, it is certain that quadrivalents are formed in a greater number at pachytene than survive until metaphase. In later stages chiasmata are found only on both sides of the centromeres. If we suppose, in accordance with DARLINGTON, that chiasmata arise at the same place as they are found at metaphase I the following possibilities are valid within each four-group of homologous chromosomes (see Fig. 3, where each pachytene chromosome is represented by a thickly drawn central portion corresponding to the zone of effective pairing, and thinner end-portions, where chiasmata are formed only in exceptional cases). If the exchange of partners can occur anywhere within different parts of the chromosomes, it is evident that the commonest case will be Fig. 3 *a,* where the exchange occurs outside the central portion. This leads to the formation of 2 typical, localised bivalents, which in earlier stages are often joined by a torsion pairing, but which are free at metaphase I. If the exchange of partners takes place close enough to the central zone (Fig. 3 *b*), it is probable that the exchange will interfere with the pairing of the central zone on that side of the centromere, so that chiasmata can be formed only on the other side of the centromere. The result will be 2 bivalents with one chiasma each. This is probably the cause of the relatively high frequency of this kind of bivalents in *Allium Porrum,* as compared with diploid species with localised chiasmata. If in the former case the pairing is inhibited only in one pair of threads there will originate a localised chain quadrivalent with 3 chiasmata (Fig. 3 *c*). Finally, if the exchange of partners occurs exactly at the centromeres, so that chiasmata can be formed on both sides of the exchange, a localised ring quadrivalent will result. It is quite comprehensible that this last case must be very rare, since it depends for its realisation on the fulfilment of quite special conditions.

SUMMARY.

Meiosis is examined in *Allium Porrum,* an autotetraploid species with complete localisation of chiasmata. Quadrivalents are formed

rather frequently at zygotene, but most of them disappear before metaphase I is reached. Chiasmata are formed only in the neighbourhood of the centromeres. A new type of metaphase quadrivalents is described: chains and rings with localised chiasmata.

Svalöf, January 20th, 1940.

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THE MODE OF CHROMOSOME PAIRING IN PHLEUM TWINS WITH 63 CHROMOSOMES AND ITS CYTOGENETIC CONSEQUENCES

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I. INTRODUCTION.

AS described in previous publications (MÜNTZING, 1937 a, 1938 a), twin seedlings with deviating chromosome numbers were obtained in 13 species belonging to 11 different genera. This material also included *Phleum pratense* L., which was represented by six triploid and three haploid aberrants. Instead of the normal number, 2n = 42, these plants had 2n = 63 and 21 respectively. For the sake of simplicity the aberrants will be referred to in the following as triploids and haploids instead of using the more adequate terms enneaploid and triploid, the normal plants of the species in reality being hexaploid (2n = 42).

The present paper is restricted to the results of a study of aberrants with 2n = 63 and their progeny. Some preliminary data concerning this work have already been given (MÜNTZING, 1937 a, 1938 b and ÅKERMAN, GRANHALL, NILSSON-LEISSNER, MÜNTZING and TEDIN, 1938), but the results now accumulated justify a fuller account.

The experimental work and the somatic chromosome counts were performed at Svalöf in the years 1935—1939 under the guidance of the senior author. The meiotic studies were undertaken at Lund after the arrival of the junior author[1]. Last year the triploid twins and their progenies were handed over to Dr. A. LEVAN, now head of the Cytogenetic Department of the Svalöf Plant Breeding Institute. The haploids will be studied and described by Miss H. NORDENSKIÖLD.

II. THE PROPERTIES OF THE TRIPLOID TWINS.

1. MORPHOLOGY AND FERTILITY.

Habitually the triploid twin plants were rather similar to their diploid sister twins, but a more detailed study revealed certain typical

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[1] At present Fellow of the Rockefeller Foundation.

differences. In one of the three twin pairs studied the triploid member was distinctly more vigorous than the diploid sister plant (Fig. 1), in the second pair both members had about the same vigour, and in the third pair the triploid plant seemed to be inferior in vigour in comparison with the diploid sister individual. Small clones were made of

Iig 1 A twin pair of *Phleum pratense* The plant to the left is triploid (2n = 63) and slightly more vigorous than the diploid sister twin to the right (2n = 42)

the six individuals, and in 1938 the average plant weight in these clones (the sum of two cuttings) was found to be the following.

Field number	Chromosome number	Number of clone plants	Average plant weight
4198 *b*	42	8	224 gr.
a	63	7	240 »
2041—2 *a*	42	4	88 »
b	63	4	103 »
4039 *a*	42	4	148 »
b	63	4	131 »

The differences between the twin pairs are evidently much greater than the differences within each pair. On an average, however, the triploid twins seem to be equal or even superior to the diploid normal plants, but more data are needed in order to prove that definitely.

TABLE 1. *Morphological data from twin clones of Phleum pratense* [1].

Field number	Chromosome number	Leaf breadth	Leaf length	Thickness of 10 leaves	Stem diameter	Culm breadth	Culm length	Spikelet length
4198 *b* ...	42	6,19 (16)	31,6 (16)	2,24 (8)	1,10 (24)	8,08 (24)	9,71 (24)	4,13 (24)
» *a* ...	63	6,93 (14)	33,0 (14)	2,66 (7)	1,31 (21)	9,17 (21)	10,10 (21)	4,62 (21)
2041—2 *a*	42	6,25 (8)	29,0 (8)	2,03 (4)	0,94 (12)	6,40 (12)	8,00 (12)	4,00 (12)
» —2 *b*	63	7,63 (8)	29,3 (8)	2,65 (4)	1,16 (12)	7,72 (12)	10,00 (12)	4,00 (12)
4039 *a* ...	42	6,63 (8)	26,0 (8)	2,23 (4)	1,18 (12)	7,72 (12)	10,00 (12)	4,50 (12)
» *b* ...	63	8,25 (8)	31,4 (8)	2 65 (4)	1,30 (12)	8,71 (12)	12,08 (12)	5,75 (12)

The diploid and triploid twins were further compared by a series of measurements, the results of which are given in Table 1. According

Fig. 2. Culms of the twin pair represented in Fig. 1. The culms of the triploid (to the right) are somewhat bigger than those of the diploid (to the left).

to these data the triploids have longer, broader and thicker leaves than the corresponding diploids and are also characterized by thicker stems,

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[1] The number of measurements for each average value between brackets For leaf and culm length the units are cm , for the other characters mm.

longer and thicker culms and bigger spikelets. The typical differences in culm and spikelet dimensions may also be seen from Figs. 2—3.

Though only three diploid-triploid twin pairs were available for comparison, the differences observed are most probably typical of material of this kind. Rather similar results were also obtained from measurements of twin clones in *Poa pratensis* (MÜNTZING, 1940). The larger dimensions of the triploid *Phleum* plants are indeed to be ex-

Fig. 3.   Spikelets of triploid *P. pratense* (upper row) and of the diploid sister twin (lower row). The dimensions of the former are somewhat larger.

pected by analogy with similar changes in numerous cases of auto-polyploidy previously studied (cf. MÜNTZING, 1936, 1937 b).

Much to our surprise pollen fertility in the triploid members of the twin pairs proved to be just as good as in the corresponding diploids, the percentage of good pollen being almost 100 per cent in all the plants examined. The appearance of pollen samples from one of the twin pairs is evident from Figs. 4—5. The only difference to be seen is a difference in size, the pollen grains of the triploid being slightly larger

than those of the corresponding diploid. The same size differences were also observed in the other two twin pairs. Measurements were undertaken in one of the pairs, the following result being obtained:

	Pollen diameter							n	M ± m	σ^2
	10	11	12	13	14	15	16			
Diploid (4039 a) ..	1	5	25	92	34	4		161	13,03 ± 0,06	0,65
Triploid (4039 b) ..				26	39	70	13	148	14,47 ± 0,07	0,78

The average values are evidently significantly different. The relation between the diameters is 14,47 . 13,03 = 1,111, which would
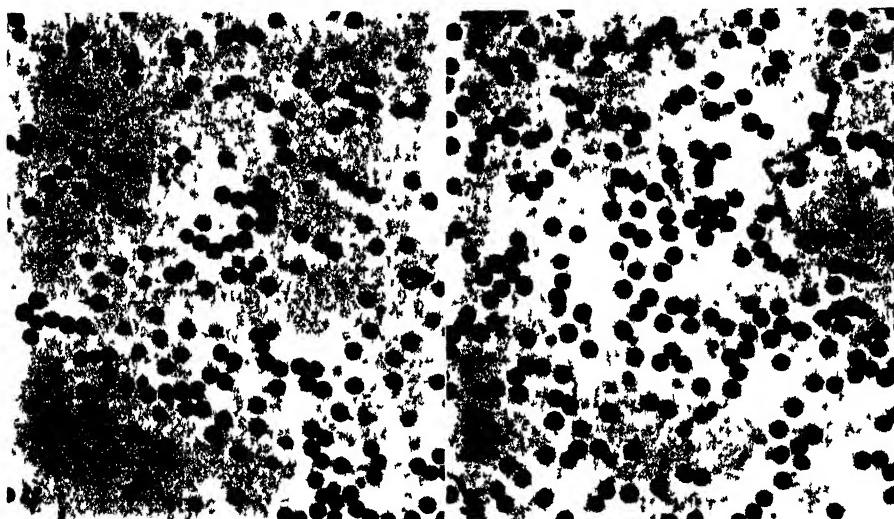
Figs 4—5 Pollen samples from a diploid—triploid twin pair of *P pratense* In both twins pollen fertility is quite good but in the triploid (Fig 5) the pollen grains are larger than in the diploid (Fig 4)

correspond to a relation of 1,111³ or 1,37 between the average volumes. The relation between the chromosome numbers of the mother plants is 63 . 42 = 1,50. Owing to elimination of chromosomes at meiosis (cf. below) the relation between the average chromosome numbers of the two categories of pollen grains will be somewhat lower than 1,50. Thus, in the present material there seems to be a rather close correspondance between chromosome number and pollen grain size.

Pollen size in the triploid seems to be slightly more variable than in the diploid, the variance values being 0,78 and 0,65 respectively. However, the odds that this difference in variability is significant are only

about 5 : 1, as may be controlled by the tables of FISHER and YATES (1938).

A greater variability in size of the pollen grains produced by the triploid was expected, since these pollen grains must contain a variable number of chromosomes. It seemed remarkable that the possible difference in variability was not much greater. However, an explanation of the unexpectedly uniform and good pollen of the triploids was obtained from a study of meiosis in this material.

Before passing on to a description of meiosis, it should only be mentioned that fertility on the female side seems to be just as good as on that of the male. No exact data on seed setting in the triploid twins are yet available, but judging from mere observations seed production in the triploids is as good as in the diploids. At any rate, much seed was obtained from crosses between different triploids, and also selfing gave a fairly good result.

2. MEIOSIS.

Meiosis was studied in the p. m. c. The material was fixed in chrome-acetic-formalin and stained with gentian violet. Though not ideal, the fixations proved to be sufficiently good to disclose the main features of meiosis.

Since the twins with 63 chromosomes are autotriploid in relation to the sister twins with the normal number 42, meiosis was expected to be rather irregular and to be characterized by a high number of trivalents and univalents. However, contrary to expectation, meiosis in the triploids was found to be remarkably regular, the number of trivalents and univalents being low and the number of bivalents high. This was evident from a study of diakinesis and first metaphase (Figs. 6—15).

Fig. 6 represents a diakinesis from the diploid twin. The 42 chromosomes appear as 21 bivalents. In most of the bivalents the chromosomes are joined by two or more chiasmata, but in some bivalents there is only one chiasma. There is no evidence that the chiasmata are localized.

In the triploid twin studied (4198 a) attempts were made to analyse a number of complete chromosome complements at diakinesis. This could be done more or less accurately in seven cells, but due to the high number of chromosomes and a somewhat unsatisfactory fixation, there were a few obscure points in each cell. However, it was immediately ·obvious that most of the chromosomes were united to bi-

valents. In addition to these there were a few univalents and some quite rare trivalents. Two such nuclei are represented in Figs. 7—8.

In Fig. 7 the chromosome complement probably consists of $2_{III} + 27_{II} + 3_I$. The trivalents are indicated by single-headed arrows.
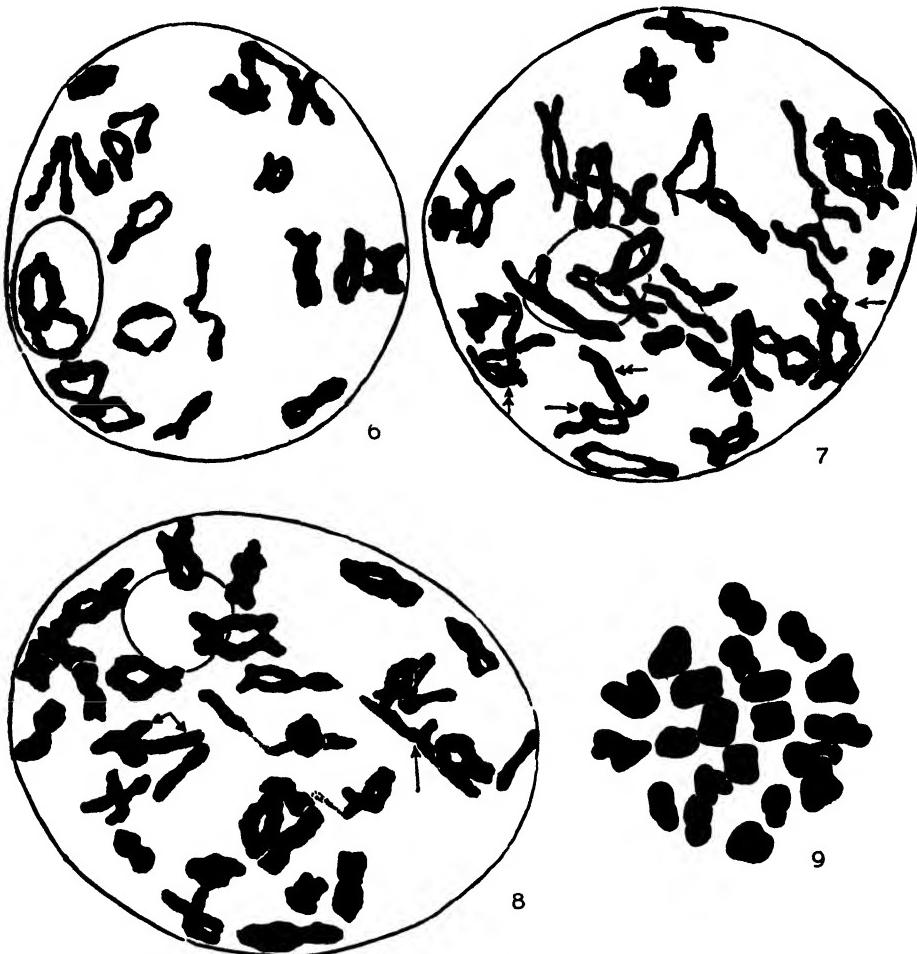
Fig. 6. Diakinesis in diploid *Phleum pratense* ($2n = 42$), 21_{II}. Figs. 7—8. Diakinesis in triploid *Phleum pratense* ($2n = 63$). Fig. 7, probably $2_{III} + 27_{II} + 3_I$; Fig. 8, probably $29_{II} + 5_I$. Fig. 9, I—M (polar view) of diploid *P. pratense*, 21_{II}. — 4170 ✕.

One of them is rather clear, the other one is somewhat more dubious. The clear one is separately drawn in Fig. 15 (the second configuration from the left). Of the 27 bivalents 24 are quite distinct and unquestionable. Of the remaining three, two (indicated by double-headed arrows)
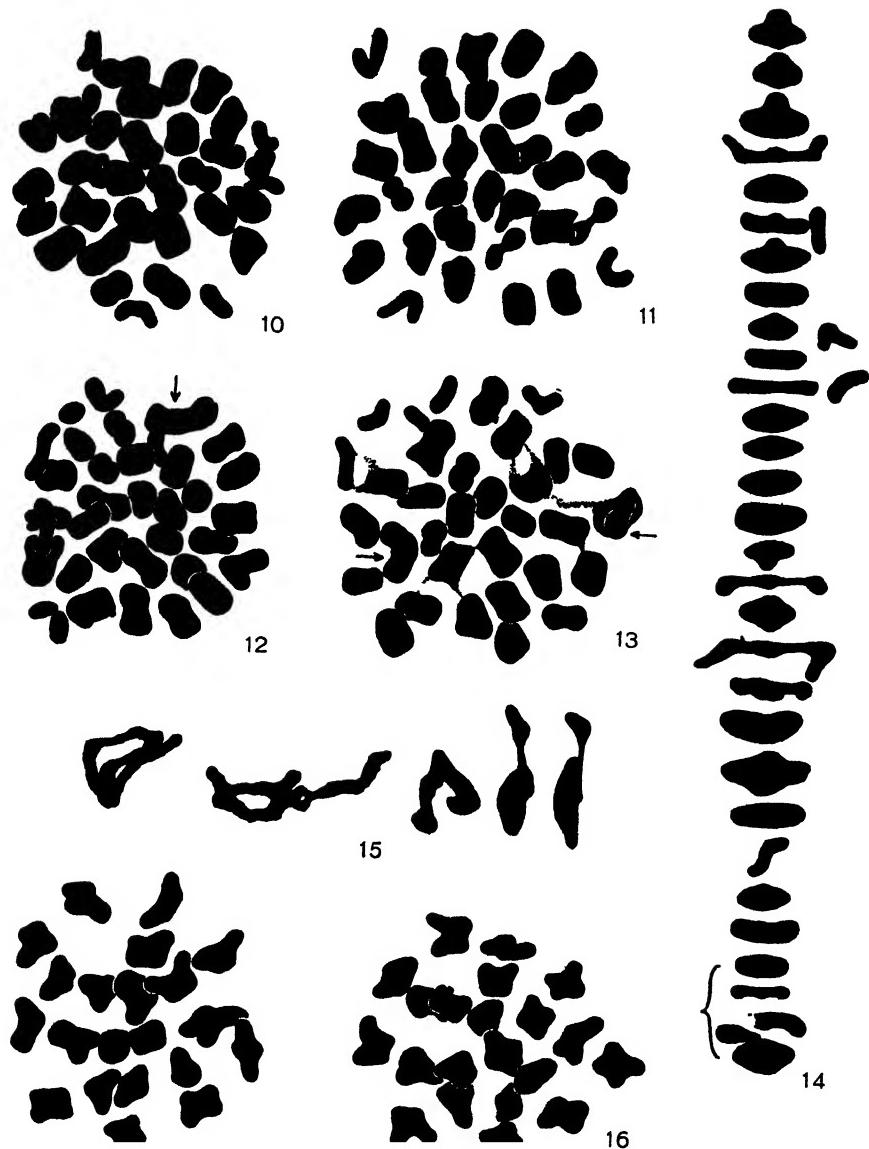
Figs. 10—14. I—M groups in triploid *Phleum pratense*. Fig. 10, $28_{II} + 7_I$; Fig. 11, probably $30_{II} + 3_I$; Fig. 12, probably $1_{III} + 27_{II} + 6_I$; Fig. 13, probably $2_{III} + 26_{II} + 5_I$; Fig. 14, side view (separately drawn), probably $1_{III} + 28_{II} + 4_I$ (the bracket indicates some not quite clear elements). Fig. 15, five separate trivalents from diakinesis (the two to the left) and first metaphase (the three to the right). Fig. 16, I—A in diploid *P. pratense* (polar view), distribution 21—21. — Fig. 14 and the three I—M trivalents in Fig. 15 drawn at a magnification of 3470 ✕, the other figures at ✕ 4170.

had not quite clearly visible contours, and in one case (the bivalent just to the right of the nucleolus) the presence of a chiasma was not quite certain. However, judging from the shape and position of the chromosomes, they most probably constitute a bivalent and not two univalents. Of the three univalents one (at 2 o'clock) is quite free, but the other two (at 8 o'clock near the nucleolus) form a cross. The arms of this cross, however, do not seem to be connected.

Since the true configurations at diakinesis are of importance, a second cell is represented in Fig. 8. The probable configuration in this cell is $29_{II} + 5_I$. No certain trivalent could be distinguished. 27 bivalents are quite clear but at two points (indicated by arrows) the associations are somewhat obscure. The arrow at 3 o'clock points to a probable chiasma, but it is not excluded that the two chromosomes represent 2 univalents instead of a bivalent. The other (double) arrow indicates a chromosome complex which has been counted as $1_{II} + 2_I$, though other interpretations may be possible. The grey connection between the two univalents in the centre is probably an artefact.

In the same way 5 other diakinesis configurations from the same plant were analysed more or less successfully. In these additional cells the following probable configurations could be distinguished: Cell 1, $2_{III} + 27_{II} + 3_I$ (possibly only 1_{III}); Cell 2, $27_{II} + 7_I$; Cell 3, $24_{II} + 5_I +$ unclear group with $\pm 3_{II}$; Cell 4, $26_{II} + 4_I +$ an unclear group; Cell 5, $28_{II} + 4_I$.

In the last four cells all the 63 chromosomes could not be distinguished, but nevertheless the data are mentioned in order to show that in all cells the number of clear bivalents was higher than 21, the number of univalents never higher than 7 and the frequency of trivalents very low.

The results obtained from the study of diakinesis were verified by observations of *first metaphase* (Figs. 9—15). In the normal twin with 42 chromosomes 21 bivalents were found to be present at I—M (Fig. 9). In the triploid *the sum of the bivalents and the occasional trivalents was hardly ever found to be lower than 28 and the number of univalents was never higher than seven*. This is evident from the following figures.

Fig. 10 shows a I—M in polar view with $28_{II} + 7_I$. The univalents are scattered round the bivalents and may be distinguished rather easily. In another I—M group (Fig. 11) the number of bivalents was higher, the number of univalents being correspondingly lower. The probable configuration in this cell is $30_{II} + 3_I$. In other cells the

presence of one or two trivalents was rather clear. In Fig. 12 the configuration is probably $1_{III} + 27_{II} + 6_I$. However, the trivalent (indicated by an arrow) was not quite certain. One of the univalents has a rather marked median constriction. — In the metaphase group represented by Fig. 13, finally, the probable configuration is $2_{III} + 26_{II} + 5_I$, but the trivalents are somewhat uncertain. The greyish streaks connecting some of the chromosomes are probably artefacts.

The chromosome configurations at I—M are also illustrated by Fig. 14, showing the result of an attempt to analyse a complete chromosome complement in side view. 25 bivalents and 4 univalents could be distinguished quite clearly. In addition to these were some additional less clear bivalents and one probable trivalent. Though all the chromosomes could not be seen with accuracy, it is nevertheless evident that the great majority of the chromosomes are present as typical bivalents. A few of them are rod-shaped, but most of them have two or more chiasmata. Though trivalents seem to be quite rare at I—M, a few associations of this kind were really observed (Fig. 15, to the right).

The figures discussed above represent only a small part of the evidence gathered. The total data obtained may be summarized in the following way.

In the diploid twin ($2n = 42$) 3 cells in polar view were found to contain 21 bivalents. In 4 cells in side view 20—21 bivalents could also be distinguished, and there was no evidence of larger associations than bivalents. Of 80 bivalents observed in side view 28 (i. e. $35{,}0 \pm 5{,}3$ per cent) were rod-shaped, the majority being ring-shaped with 2 or more chiasmata [1].

In the triploid ($2n = 63$) attempts were made to analyse 5 different I—M groups in side view, but in none of them could the complete chromosome complement be distinguished. However, the following numbers of bivalents were visible: 27, 25, 23, 28 and 27 respectively. In the same cells the number of univalents observed varied between 4 and 7. A total of 4 more or less clear trivalents were also observed, no cell containing more than one trivalent.

The proportion of rod-shaped bivalents was found to be about the same as in the normal twin. Of 130 bivalents studied 38 belonged to this category, the others being rings with at least 2 chiasmata. This gives

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[1] It is possible that the true proportion of ring bivalents is slightly higher. The value mentioned is based on counts in a few, particularly clear I—M groups, in which the proportion of rod bivalents may be somewhat higher than the average. The same is true of the corresponding counts in the triploid.

a percentage value of 29,2 ± 4,0. The difference between this value and the corresponding percentage of the diploid 35,0 ± 5,3 is not significant. At any rate, the triploid is *not* characterized by a higher frequency of rod bivalents than the diploid.

In the triploid a total of 11 cells in polar view were studied. In seven of these the total number of chromosomes was found to be 63. These cells represented the following configurations: $28_{II} + 7_I$ (3 cells), $1_{III} + 27_{II} + 6_I$ (2 cells); $2_{III} + 26_{II} + 5_I$ (1 cell) and $30_{II} + 3_I$ (1 cell). In the remaining 4 cells one chromosome was lacking, the total number of chromosomes distinguished being 62. Though these configurations are incomplete the configurations observed may be mentioned, viz. $28_{II} + 6_I$ (2 cells); $1_{III} + 27_{II} + 5_I$ (1 cell) and $1_{III} + 26_{II} + 7_I$ (1 cell). In the last-mentioned cell the sum of the bivalents and trivalents is most probably not higher than 27. This is the only exception so far observed to the rule that the sum of bivalents and trivalents in this material is not lower than 28.

The more or less successful analysis of entire I—M groups was supplemented by counts of the number of univalents per cell. Counts in four different slides gave the following total results:

	Number of univalents						Number of cells	M ± m
	2	3	4	5	6	7		
Polar view...		2	4	7	10	6	29	5,48 ± 0,22
Side view	2	9	15	30	24	9	89	5,03 ± 0,12

Since some univalents may have been overlooked, the numbers counted represent minimum values. However, the difference between the true and the calculated average values must be relatively slight, the univalents generally lying more or less peripherically and thus being easy to distinguish. At any rate, the true average value is lower than 7, some cells certainly having less than 7 univalents. It is still more important, however, that not a single one of the 118 cells studied was found to contain more than 7 univalents.

When comparing diakinesis and first metaphase the frequency of univalents seems to be somewhat higher at the latter stage. In seven diakinesis cells the probable number of univalents varied from 3 to 7 with an average of 4,43. On inspection of a larger number of cells at diakinesis the impression was obtained that the number of univalents at this stage was in fact slightly lower than at first metaphase.
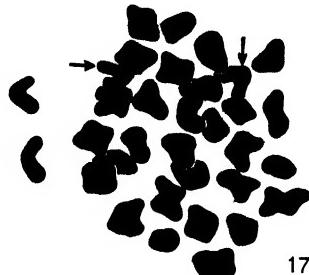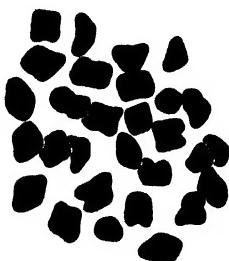
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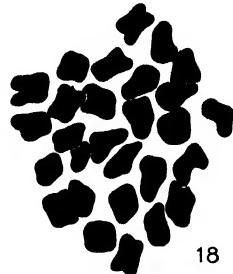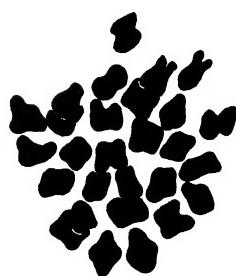
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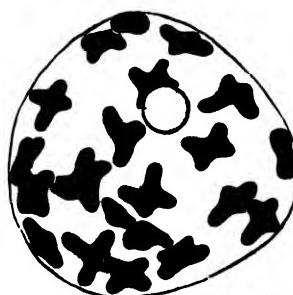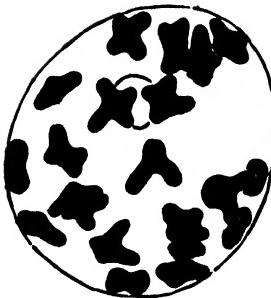
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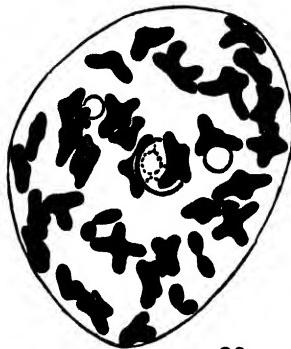
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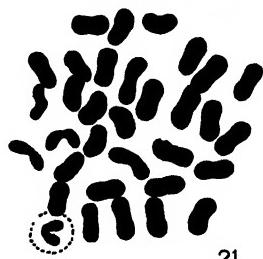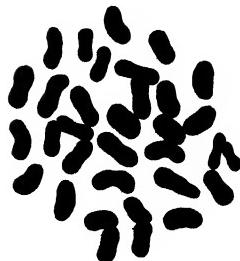
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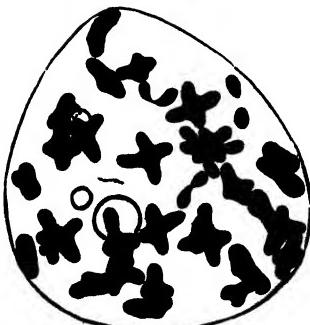
First anaphase is quite regular in the diploid, 21 chromosomes going to each pole. In some cells the position of the chromosomes was found to be so regular that the corresponding chromosomes in the two anaphase groups could be distinguished without difficulty (Fig. 16). First anaphase in the triploid (Figs. 17—18) is also rather regular, though minor complications arise due to the presence of univalents. Most of the univalents present at I—M lag and split at I—A, but some of them, lying far from the equator, pass to the poles undivided. The division of the univalents takes place at late anaphase and has not yet occurred in the cells represented by Figs. 17—18. In Fig. 17 the I—M configuration has evidently been $29_{II} + 5_I$ (or possibly $2_{III} + 26_{II} + 5_I$). At I—A three of the univalents are lagging, but the remaining two (indicated by arrows) are situated below the lower of the anaphase groups and would certainly be included in the interphase nucleus without division. The 29 chromosomes in the two anaphase groups have a corresponding position, which makes it possible to decide rather accurately which chromosomes have been united as bivalents at I—M. The same corresponding position may also be seen rather well in the anaphase chromosomes of Fig. 18. In this cell the two anaphase groups contain 28 chromosomes, 7 chromosomes being situated between the groups. All these 7 chromosomes are probably former I—M univalents, though the possibility is not quite excluded that some of them might be delayed members of previous bi- or trivalents. However, the strict correspondance of the members in the two anaphase groups represents evidence against such an interpretation.

The number of univalents dividing at late I—A was counted in four slides with the following total result:

Number of dividing univalents:	0	1	2	3	4	5	6	7	n	$M \pm m$
» » cells:	2	4	8	8	12	9	4	2	49	$3{,}57 \pm 0{,}25$

The average number of dividing univalents is evidently significantly lower than the number of univalents present at I—M ($5{,}48 \pm 0{,}22$ in

Figs. 17—18, I—A in triploid *Phleum pratense* (polar view). Fig 17, distribution 29—3—31 (two elements in the right group, indicated by arrows, are undivided univalents lying at a deep level). Fig. 18, distribution 28—7—28. — Fig. 19, early interphase in diploid *P. pratense,* 21 chromosomes in each nucleus. Fig. 20, early interphase in triploid *P. pratense.* (In the lower nucleus 29 chromosomes + 5 half univalents, in the upper nucleus 30 chromosomes + 3 half univalents.) Fig. 21, II—M in triploid *P. pratense.* (In the left group probably 31 chromosomes + 2 half univalents; near the right group one eliminated half univalent; in the right group probably 29 chromosomes + 3 half univalents.) — $\times$ 4170.

polar view and 5,03 ± 0,12 in side view). This confirms the observation that a proportion of the univalents pass to the poles without division.

The fate of the univalents and the chromosome distribution could be further studied at *interphase*, the chromosomes at the early part of this stage being clearly visible (Figs. 19—20). A typical interphase from the triploid is represented by Fig. 20, which may be compared with the corresponding stage in the diploid (Fig. 19). In Fig. 19 there are, as expected, 21 chromosomes in each cell, in Fig. 20 the situation is more complicated. In the lower nucleus there are 29 ordinary chromosomes and probably 5 half univalents (one of these apparently divided into two parts, due to the pronounced median constriction). In the other nucleus there are 30 ordinary chromosomes and 3 half univalents. This result shows that the two halves of a split univalent may sometimes be included in the same interphase nucleus.

At interphase the proportion of eliminated chromosomes was found to be quite low. Thus, even the products of the lagging and splitting univalents are in most cases included in the nuclei. In 78 cells observed, the number of eliminated chromatids was found to be the following:

Number of eliminated chromatids:	0	1	2	3	n	M
» » p. m. c.:	52	11	13	2	78	0,55

The eliminated chromatids were generally present as half univalents, but in some cases the two chromatids of a univalent had not separated. In most of the pollen mother cells, however, there was no chromosome elimination at this stage, the average number of eliminated chromatids being as low as 0,55.

At interphase the chromosome distribution was studied in ten cells, including the one described above (Fig. 20). In six of these cells it was possible with more or less accuracy to distinguish the split univalents from the other chromosomes. In these cells the distribution was the following: (29 + 5/2) — (30 + 3/2) (Fig. 20); (29 + 4/2) — (31 + 2/2); (28 + 5/2) — (31 + 3/2); (30 + 2/2) — (31 + 2/2); (29 + 2/2) — 2/2 eliminated — (32 + 0); (29 + 2/2) — 2/2 eliminated — (30 + 2/2). In the last-mentioned cell a total of only 62 chromosomes could be distinguished. In 4 additional cells the split univalents could not with certainty be distinguished from the other chromosomes. The number of elements in the two nuclei of these p. m. c. were 32—33, 32—35, 31—32 and 34—35 respectively. Since the total number of elements in these cells varies from 63 to 69, this represents additional evidence that

the univalents splitting at I—A are often included in the interphase nuclei.

At *second metaphase* the split univalents could be distinguished rather well from the other chromosomes. In Fig. 21 the metaphase group to the left most probably contains 31 ordinary chromosomes + 2 half univalents (at 3 o'clock). The other group seems to be composed of 29 chromosomes + 3 half univalents. One half univalent is eliminated. Thus, in this case 3 univalents have evidently divided at first anaphase.

The number of half univalents lying more or less scattered outside the II—M plates was counted in 22 p. m. c. The number of such elements per II—M plate was found to vary between 0 and 5 with an average of 2,32. Since the average number of univalents dividing at I—A was found to be 3,57 the majority of the split univalents evidently lie more or less scattered round the II—M plates.

At *second anaphase* lagging chromosomes were frequent, these laggards undoubtedly being represented by the split univalents. The number of lagging chromosomes was counted in 50 cells, the result being the following:

Number of lagging chromosomes:	1	2	3	4	5	6	n	M
» » cells:...................	4	11	8	14	8	5	50	3,52

The average number of lagging chromosomes closely corresponds to the average number of univalents dividing at I—A, the two values being 3,52 and 3,57 respectively. Thus, it is highly probable, as is usual in such cases, that univalents dividing at I—A and being included in the daughter nuclei appear as laggards at II—A. A small proportion of the split univalents is eliminated already in the first division, but most of the elimination evidently takes place at II—A.

An attempt was made to estimate the degree of elimination by observations of the number of eliminated chromosomes in young tetrads. These eliminated chromosomes were in a few cases lying in clear micronuclei but generally apparently free in the plasm. In most cases the eliminated bodies evidently correspond to one chromatid (= half a univalent), but sometimes apparently to two chromatids. In a few cases the chromosomal nature of the eliminated bodies was doubtful. Even if, for these reasons, the counts are not quite accurate, they nevertheless represent the true situation fairly well. The following numbers were found:

Number of eliminated chromatids:	0	1	2	3	4	5	n	M
» » tetrad cells:	34	50	28	24	7	1	144	1,47

The average value allows a calculation of the probable average number of chromosomes in the male gametes of the triploid plants. This would be $63/2 - 1{,}47 = 30{,}03$. If the same degree of elimination occurs in the ovules, and if there is no selective zygotic elimination, the average chromosome number of the offspring would be 60,06. At any rate, owing to the observed elimination of chromosomes, the average chromosome number in the offspring should be lower than 63. It might also be predicted that in the progenies of the triploid twins the chromosome number should only in very exceptional cases be lower than 56, practically all gametes receiving at least 28 chromosomes. As described below these predictions were indeed verified.

III. PROGENIES OF TRIPLOID TWINS.

1. MATERIAL.

In 1936 the three twins with 63 chromosomes (Nos. 4198 a, 2041— 2 b and 4039 b) flowered for the first time. The culms produced were in part isolated, in part cross-pollinated. Seed after isolation was obtained from 4198 a and 4039 b and after crossing from the combinations 4198 $a \times$ 4039 b and reciprocally, 4039 $b \times$ 2041—2 b and 2041—2 $b \times$ $\times$ 4198 a. The seeds obtained were quite good and were observed to be larger than ordinary timothy seeds. They were germinated in April, 1937 on a germination apparatus, the seedlings were planted in boxes with sterilized soil and later on transplanted to the field. Germination was quite good, the percentage values being 99 for the seed obtained after isolation (122 seedlings from 123 seeds) as well as for the cross-pollinated seed (1589 seedlings from 1611 seeds). As a standard seed of the commercial variety »Gloria» ($2n = 42$) was germinated at the same time. The percentage of germination of the standard was 98. In the field the plants were planted in rows with the distances 40 cm. between the plants and 60 cm. between the rows. Every 10th plant in the rows was a standard plant, thus allowing a rather good comparison between standard and the material to be tested.

2. CHROMOSOMAL VARIATION.

Since the offspring of the triploid plants was represented by as many as 1711 seedlings, it was not possible to determine the somatic chromosome number in all this material. However, a rather good idea of the chromosomal variation was gathered from counts in a total of

186 individuals.   The majority of the fixations (148) were made at the seedling stage, care being taken to avoid selection of special seedlings. Later on 38 fixations were also made from especially vigorous plants in the field.   The results of the chromosome counts are given in Table 2.

The main result of the chromosome counts is the fact that *of the 186 individuals tested not a single one had a lower chromosome number than 56*.   This is a verification of the meiotic observation that practically all gametes receive at least 28 chromosomes.   The kind of chromosomal variation in the offspring is also in harmony with the observed mode of meiosis.   The numbers found range from 56 to 64, thus most of the

TABLE 2.   *Chromosome numbers in the offspring of triploid Phleum pratense* $(2n = 63)$.

Field number		Somatic chromosome number									n	M	σ^2
		56	57	58	59	60	61	62	63	64			
37—2....................	Without selection	3	1	6	10	8	7	1	3	1	40	59,63	
—3....................		2	2	5	10	8	9	2	2	4	44	60,02	
—4....................		1	2	2	5	6	3	2	5	1	27	60,15	
—5....................		4	3	6	8	11	3	1	1		37	59,00	
Total		10	8	19	33	33	22	6	11	6	148	59,73	3,87
37—1....................	Selected vigorous plants	3	2	1	—	1					7	57,14	
—2....................			1	1							2	57,50	
—3....................		1	—	2	6	2	3	1	4	2	21	60,52	
—5....................			1	2	—	2	—	1			6	59,17	
—6....................						1	—	1			2	60,00	
Total		4	4	6	7	5	4	2	4	2	38	59,50	5,45
Total of all values		14	12	25	40	38	26	8	15	8	186	59,66	

plants had numbers lower than those of the mother plants. A calculation of percentage values gives 87,6 per cent with lower numbers than the mother plants, 8,1 per cent with the same number and only 4,3 per cent with a higher number.

All the progenies are similar in this respect, the progenies raised after isolation behaving in the same way as those obtained after crossing. The same range of chromosome variation was also found among the selected vigorous plants as among those taken at random.

The total average chromosome number in the progenies is 59,66, this value being 3,34 lower than 63, the chromosome number of the mother plants.   The average decrease is certainly due to the chromosomal elimination occurring at meiosis in the mother plants.   According

to the meiotic data given above (p. 478) the average chromosome number of the gametes was calculated to be 30,03. The union of such gametes would result in an average chromosome number of 60,06. The agreement between expectation (60,06) and observation (59,66) is evidently quite good.

For control a few chromosome counts were also undertaken in the standard variety »Gloria». Eight plants were examined and, as expected, found to have 2n = 42, the normal chromosome number of *Phleum pratense*.

3. PLANT WEIGHT.

The material available allowed a rather accurate comparison between the progenies of twin plants with 2n = 63 and the commercial

TABLE 3. *Plant weight in the offspring of Phleum pratense twins with 63 chromosomes.*

Field number	Sum of 4 weighings (in hg.)											n	M ± m (in gr.)	σ²
	0	2	4	6	8	10	12	14	16	18	20			
⎰ 37—1	8	32	52	34	25	11	12	1	3			178	658 ± 24	2,89
⎱ standard		3	2	4	5	3	2	1				20	830 ± 70	2,98
⎰ 37—2	2	2	9	24	22	9	6	3	1			78	844 ± 36	2,33
⎱ standard		2	1	1	5	4	1					14	858 ± 84	2,34
⎰ 37—3	10	33	80	155	138	115	36	13	7			587	832 ± 14	2,81
⎱ standard		7	7	18	11	10	12	2	—		1	68	876 ± 38	3,12
⎰ 37—4	2	7	13	6	3							31	506 ± 56	1,10
⎱ standard				1	—	2	—	1				4	1100 ± 157	2,67
⎰ 37—5	12	31	60	113	108	75	39	9	3			450	820 ± 15	2,47
⎱ standard		10	6	10	5	13	5	3	1		1	54	860 ± 43	4,20
⎰ 37—6	2	12	18	12	3							47	508 ± 46	0,95
⎱ standard			1	1	2	—	1					5	860 ± 140	2,20
37—1→6, total	36	117	232	344	299	210	93	26	14			1371	787 ± 8	2,56
standard, total		22	17	35	28	32	21	7	1		2	165	867 ± 25	3,30

variety »Gloria», having the normal chromosome number 42. The first thing to be studied was vigour and productivity as measured by plant weight. The field plants were cut and weighed four times (once in 1937, twice in 1938 and once in 1939). The sum of these weighings may be considered to be a good expression of vigour in this material. The results obtained are summarized in Table 3.

The first fact to be seen from the table is that the total yield of the

six families is less good than that of the standard, the average values being 787 ± 8 and 867 ± 25 gr. respectively. The difference is significant (D/m = 3,12). However, among the six families there are considerable differences, some of them having about the same average value as the standard, others being quite inferior. The best families are those involving the mother plant 4198 *a*, which had previously been observed to be much more vigorous than the other two twins with 2n = 63 (4039 *b* and 2041—2 b). Progeny 2 is the offspring of isolation of 4198 *a*, and though timothy is a plant species which is rather sensitive to inbreeding (SYLVÉN, 1929; VALLE, 1931), the average value 844 ± 36 is almost as high as that of the corresponding standard, 858 ± 84. The difference is not significant. In three other progenies, 37—1, 37—3, and 37—5, the mother plant 4198 *a* is also involved. Two of these represent crosses 4198 *a* × 4039 *b* (37—3) and reciprocally (37—5), and in these progenies the average values are quite good and not significantly lower than those of the standard. In the third family (37—1) the average vigour is rather poor and most probably lower than in the standard (D/m = 2,32). This family is derived from the cross 2041—2 *b* × × 4198 *a*. Evidently this cross combination is rather unfavourable, the plant 2041—2 *b* being rather poor. It is also possible that a considerable proportion of the plants are the result of self-fertilization, the self-sterility of the mother plants (in this case 2041—2 *b*) not being complete. The two remaining families (37—4 and 37—6) are very poor, and this is not surprising, since they do not involve the vigorous triploid twin 4198 *a*. 37—4 is derived from isolation of 4039 *b*, and 37—6 represents the cross 4039 *b* × 2041—2 *b*.

In spite of the fact that there is a good deal of chromosome variation in the progenies of triploid twins, these progenies appeared just as uniform in the field as the cytologically stable standard variety. In fact, the variance values of the twin progenies were all found to be *lower* than those of the corresponding standard plants. This, however, may be due to the fact that the standard plants were growing at wider distances from each other than the other plants, thus being more exposed to the soil variation. At any rate, the variability in the twin progenies, in spite of the chromosomal diversity, was of about the same order as in the standard variety.

When comparing the vigour of the twin progenies and the standard, the different weighings were observed to give rather different results. This implies that in the beginning, when the plants were young, the twin progenies were more vigorous in relation to the standard than

later on. Thus, at the first weighing in 1937 the total average of the
twin progenies was 140,25 ± 1,70 gr., the corresponding average of the
standard, 123,15 ± 4,64 gr., being significantly lower.

However, in the following year, 1938, when the plants had wintered,
the twin progenies yielded less than the standard, and this decrease
continued also in 1939. If the standard average every time is given
the value 100, the corresponding average of the twin progenies was
114 in 1937, 97 in 1938 and 91 in 1939. These values are based on the
total yield, the value 97 in 1938 representing the sum of the yield in
1937 and 1938 and the value 91 in 1939 the sum of the weight in all
three years. This obvious change in the relation between twin progenies
and standard is probably due in the first place to a difference in winter
hardiness. Further, the high weight of the twin progenies in the first
summer may be connected with the fact that the seedlings of this
material get a better start than the seedlings of the standard. Though
germinating simultaneously with the standard, the seedlings of the twin
progenies already at an early stage were conspicuously larger than the
standard seedlings. This early difference is probably in part connected
with the observed difference in seed size, the bigger seeds giving more
vigorous seedlings. Thus, the higher chromosome number of the tri-
ploid twins is probably in this way advantageous to the early develop-
ment of the offspring, and this initial advantage may partly be
responsible for the good yield of the first crop. The good development
of the offspring in the first year must also and perhaps in the first
place be ascribed to a general effect of the increased chromosome num-
ber. Later on, however, the favourable relation between the yield of
the twin progenies and the standard is disturbed by the insufficient
average hardiness of the former.

4. CORRELATION BETWEEN CHROMOSOME NUMBER AND VIGOUR.

As the twin progenies consisted of plants with at least nine differ-
ent chromosome numbers, it was desirable to test the possible occurrence
of a correlation between chromosome number and vigour. For this
study 184 individuals with known chromosome numbers were available.
Considering first the correlation between chromsome number and total
yield in the three years, the following average values were obtained in
the different chromosome classes:

Chromosome number:	56	57	58	59	60	61	62	63	64
Average weight:	894	1013	1005	848	825	889	806	845	1050
Plant number:	13	12	25	39	38	26	8	15	8

. These values do not reveal any obvious correlation. If, however, the material is concentrated to the three chromosome classes 56—58, 59—61 and 62—64 the following distribution of the variates is obtained (Table 4).

According to this table the plants having 59—61 chromosomes are probably less vigorous than those having 56—58 chromosomes, the average weight values being 849,8 ± 28,4 and 978,0 ± 49,0 respectively. The difference is 128,2 ± ± 56,6 and D/m = 2,27. The plants with the highest chromosome numbers, 62—64, also seem to be more vigorous than those having intermediate numbers, but this difference is too slight to be of any significance.

The real occurrence of a correlation between chromosome number and vigour is strengthened by the following data. It was observed that at the last weighing in 1939 the weight differences between the chromosome classes were much more pronounced than at the first weighing in 1937.

In the first weighing the absolute weight values in the three chromosome classes were 184,0, 168,7 and 175,0 respectively. In the fourth weighing the corresponding values were found to be 266,0, 212,9 and 234,7. In the latter series the difference between the average values 266,0 and 212,0 was found to be 53,1 ± 20,2. This gives a D/m value of 2,63, which strongly indicates that the intermediate chromosome class in this year was less vigorous than the 56—58 class. For comparison it may also be convenient to give the relative values in the following manner:

	Chromosome classes		
	56—58	59—61	62—64
1st weighing:	109	100	104
4th » :	125	100	110

Evidently the weight minimum in the intermediate chromosome class is much more pronounced at the fourth weighing in 1939 than

TABLE 4. *Correlation between chromosome number and vigour in the offspring of triploid Phleum pratense.*

Chromosome number	Plant weight													n	M ± m
	0	150	300	450	600	750	900	1050	1200	1350	1500	1650	1800 gr.		
56—58			1	2	3	8	7	8	10	3	3	4	1	50	978,0 ± 49,0
59—61		2	—	5	13	21	17	16	16	11	2			103	849,8 ± 28,4
62—64			1	4	—	7	4	4	6	2	1	2		31	888,0 ± 63,3

at the first weighing in 1937. This strongly indicates that the plants having 59—61 chromosomes are less resistant to frost and other adverse environmental conditions than those having exactly or approximately the euploid number 56. The plants with $\pm$ 63 chromosomes also seem to be better than the intermediate class, but this difference is not certain.

More evidence as to the occurrence of correlation between chromosome number and vigour was obtained from a comparison between the chromosome numbers of the plants taken at random and those selected on account of their high weight.

According to Table 2 the average chromosome number of the plants taken at random is approximately the same as the average chromosome number of the selected vigorous plants, the two values being 59,73 and 59,50 respectively. However, the two series seem to differ from each other in another respect, viz. the degree of variance. Among the selected plants the numbers approaching the extreme values 56 and 64 are relatively more numerous than those having intermediate chromosome numbers. This implies an increased variability in comparison with the other series, in which the variates are more concentrated in the middle. The variance values of the two series were found to be 5,45 and 3,87 respectively (Table 2). It is rather probable that the former variance is really greater than the latter, since the relation is 5,45 : 3,87 = 1,41 and P about 0,1 (according to the tables by FISHER and YATES, 1938). This result represents additional evidence that the plants having the exact or approximate 8x constitution are slightly more vigorous than those having numbers intermediate between 56 and 63. It is also possible that the plants having $\pm$ 63 chromosomes are more vigorous than the intermediate class, but the evidence for this conclusion is less convincing.

5. FERTILITY.

As in the mother plants, fertility proved to be perfectly good in the twin progenies. Since, in order to get their weight, the field plants were cut down before the seeds were ripe, an estimation of seed setting could only be made on pot plants cultivated in the greenhouse. The plants, representing the whole range of chromosome numbers, had been taken from the field in order to raise progeny after isolation or crossing. In plants having flowered in crossing groups the number of seeds per cm. of the culm was found to range from 0 to 110, the average value being 55,7. This is a degree of seed setting quite comparable to that found in ordinary timothy (cf. MÜNTZING, 1935, p. 105).

More precise information as to fertility was gathered from an examination of pollen samples, which could be taken before the plants were cut and weighed. The following results were obtained:

	Per cent good pollen						
	75—80	— 85	— 90	— 95	— 100	n	M
Plants in the twin progenies: ..	1	—	5	39	167	212	96,25
Standard plants:		1	1	3	3	8	92,50

Besides the standard plants having 80—100 per cent good pollen there was one male sterile standard individual with non-dehiscing anthers.

According to these data pollen fertility in the twin progenies is perfectly good and at least as good as in the standard variety. The pollen samples in the twin progenies were in the first place taken from the plants with known chromosome numbers. As practically all the plants were found to have 90—100 per cent good pollen, there is no correlation between pollen fertility and chromosome number. The pollen is good irrespective of the chromosome number.

IV. DISCUSSION.

The twin plants with 2n = 63 are undoubtedly autopolyploid. They develop from the same seeds as the sister twins with 2n = 42 and are most probably the result of a union between unreduced ovules and reduced male gametes (MÜNTZING, 1937 a, p. 222). In a previous paper (MÜNTZING, 1935) the genomatic constitution of *Phleum pratense* was given as *NN AA BB.* Consequently the twins with 63 chromosomes should have the formula *NNN AAA BBB.*

It is true that among the three sets of 21 chromosomes, constituting the triploid twins, there are certainly gene differences, *Phleum pratense* being a cross-fertilizing species. However, just as the two sets of 21 chromosomes in diploid *pratense* conjugate quite normally in spite of the presence of gene differences, the same good conjugation should be expected in the triploid twins between the three sets of 21 chromosomes. This would give rise to a considerable frequency of trivalents of the constitution *NNN, AAA* and *BBB,* a maximum of 21 trivalents being possible. Obviously, however, this mode of conjugation was not met with, the trivalents being quite rare and no possibly occurring larger associations being observed with certainty. At diakinesis the number of trivalents was certainly not higher than one or two, and at first

metaphase the number was evidently still lower. Only in a few I—M groups could quite clear trivalents be distinguished, and in comparison with the bivalents they were quite exceptional.

The very low frequency of trivalents must evidently be considered in connection with the unexpectedly high frequency of bivalents. In the absence of trivalents, 28 was the most frequent number of bivalents, and if the cell contained one or a few trivalents, the sum of the bivalents and trivalents was not lower than 28 [1]. The correctness of these observations was verified by chromosome counts in the offspring. Of 186 plants examined not a single one had less than 56 chromosomes.

The increased number of bivalents must be due to intergenomatic pairing between the N, A and B genomes. Since the expected number of associations is increased by seven, from 21 to 28, two of the three genomes may be supposed to be homologous. Assuming arbitrarily that these genomes are A and B, their homology is better indicated by using the symbols A_1 and A_2. Thus, the genome formula of diploid and triploid *pratense* would be $NN A_1A_1 A_2A_2$ and $NNN A_1A_1A_1 A_2A_2A_2$ respectively. In the triploid the 28 bivalents formed would consist of $NN + A_1A_1 + A_2A_2 + A_1A_2$. The remaining seven N chromosomes evidently may sometimes associate with homologous chromosomes (probably in the first place with the other N chromosomes, cf. below p. 488), giving a few trivalents, but most of them appear as univalents. In a few cases the number of bivalents or bivalents + trivalents was found to be higher than 28. This can only be explained by assuming a certain extent of intragenomatic homology. Pending more accurate data on the occurrence of more than 28 associations at diakinesis and I—M in the triploid twins, a detailed consideration of this probable intragenomatic pairing may be postponed till later on. The important thing to be discussed now is the intergenomatic pairing and the very low frequency of trivalents.

The homology between at least two of the *pratense* genomes has already been demonstrated by NORDENSKIÖLD (1937). From crosses between *Phleum pratense* (2n = 42) and *P. nodosum* (2n = 14) this author obtained two hybrid plants having 2n = 28. In these hybrids meiosis was found to be quite regular, 14_{II} being the typical I—M configuration. In a few cells a ring-shaped quadrivalent was also observed. The regular meiosis in this hybrid combination is evidently in perfect agreement with.the mode of meiosis in the triploid *pratense* twins. Using

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[1] With one possible exception (p. 473).

the same genome symbols, *Phleum nodosum* will have the constitution NN, *P. pratense* $NN\ A_1A_1\ A_2A_2$ and the F_1 hybrids $NN\ A_1A_2$. The formation of 14 bivalents in the hybrid is most probably due to conjugation of N with N and A_1 with A_2. Thus, the A_1 and A_2 genomes pair regularly just as in the triploid *pratense* twins.

Though the homology of A_1 and A_2 helps to explain the high number of bivalents in the triploid *pratense* twins, it leaves the main problem unsolved, viz. the remarkably low frequency of trivalents. This is not due to a general inability of the *Phleum pratense* and *nodosum* chromosomes to form multivalents. In the material previously studied by MÜNTZING (1935) and NORDENSKIÖLD (1937) associations of 3, 4 or even 5 chromosomes occurred in various hybrids or autopolyploid forms. This is not surprising since the chiasma frequency is, indeed, sufficiently high to permit the formation of such multivalents. In the triploid twins $(2n = 63)$ the majority of the I—M bivalents were rings with a least 2 chiasmata. At diakinesis the chiasma number in the bivalents was found to range from 1 to 3 or even 4 (Figs. 6—8). It also seems clear that the chiasmata are not localized to special regions, which would have been an obstacle to multivalent formation.

Thus, *the Phleum chromosomes are not incapable of forming multivalents, but there is a tendency for such associations to be formed only when there is no opportunity of pairing two-by-two*. In autotriploid *Phleum nodosum* $(2n = 21)$ trivalents were not rare (NORDENSKIÖLD, 1937) one metaphase group reproduced showing the configuration $4_{III} + 3_{II} + 3_I$ (l. c. Fig. 5). In the triploid *Phleum pratense* $(2n = 63)$ there are also three quite homologous N genomes besides the A_1 and A_2 genomes. Since the latter form only bivalents $(A_1—A_1,\ A_2—A_2$ and $A_1—A_2)$ the pairing of the N genomes in the triploid *pratense* should be expected to be of exactly the same kind as in the triploid *nodosum*. This is not the case, however, the frequency of trivalents evidently being much lower in triploid *pratense* than in triploid *nodosum*. This difference may be attributed to the difference in absolute chromosome number. In triploid *nodosum*, having only 21 chromosomes, the chance that the three homologous chromosomes find each other and get paired at zygotene is much greater than in triploid *pratense,* in which the chromosome number is three times as high.

In this connection the results of UPCOTT (1939 a) are interesting. The chiasma frequency of the duplex tetraploid hybrid between *Primula floribunda* and *P. verticillata* was found to be slightly lower than in the parent species. Other tetraploids, equally allo and auto, were found to

obey the same reduction rule [1]. According to UPCOTT the most probable explanation is that in the tetraploids the more numerous chromosomes take longer to pair and therefore partially fail to associate, the frequency of chiasmata being proportional to the amount of pairing.

In the pentaploid *Phleum nodosum* $\times$ *pratense* hybrids (2n = 35), studied by NORDENSKIÖLD (1937), the preliminary data demonstrate a rather high degree of multivalent formation. Two metaphase groups reproduced represent the configurations $2_{IV} + 2_{III} + 8_{II} + 5_I$ and $2_V + 2_{III} + 8_{II} + 3_I$ (l. c. Figs. 3—4). Hybrids of this kind have the constitution $NNNA_1A_2$. If chromosome pairing in such hybrids proceeded along the same lines as in triploid *pratense*, the typical configuration should be $14_{II} + 7_I$. A low frequency of trivalents might also be expected, the sum of bi- and trivalents not being higher than 14. Evidently, the degree of multivalent formation is greater than expected, and this may perhaps be attributed to the difference in absolute chromosome number, 35 versus 63. — In the pentaploid hybrid not only the homology between the N genomes and the homology between A_1 and A_2 results in pairing but also a partial homology between the N and the A genomes. In the tetraploid *pratense* $\times$ *nodosum* hybrid discussed above a few quadrivalents were also observed. These must also result from the occasional association of A and N chromosomes.

It is remarkable that in triploid *pratense* pairing between the A_1 and A_2 genomes is much more easily realized than pairing between the three A_1 genomes or the three A_2 genomes. Thus, bivalents of the type A_1—A_2 are regularly formed in spite of the fact that the A_1 and A_2 genomes might be supposed to be only partially homologous. On the other hand, the perfectly homologous associations A_1—A_1—A_1 and A_2—A_2—A_2 are very seldom realized. If the low frequency of multivalents in the plants with 2n = 63 was due solely to the high absolute chromosome number, making it difficult for the homologous chromosomes to find each other, it is hard to understand why the seven A_1—A_2 bivalents are regularly formed and not A_1—A_1—A_1 and A_2—A_2—A_2 trivalents.

The possibility is not excluded that the A_1 and A_2 genomes are in reality perfectly homologous (cf. below p. 493), but even on this assumption the mode of chromosome conjugation in triploid *pratense* is peculiar. Moreover, if A_1 and A_2 are homologous they should be expected to give

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[1] This rule, however, is not without exceptions. In autotetraploid *Dactylis glomerata* the chiasma frequency was the same as or even somewhat higher than in diploid *Dactylis Aschersoniana* (MÜNTZING, 1937 b, p. 154).

multivalents in pure *Phleum pratense*, but apart from rare exceptions such multivalents are not formed, the typical I—M configuration of *Phleum pratense* being 21_{II}.

As already pointed out above, the only possibility is to assume that the tendency to two-by-two pairing in this species is much more marked than the tendency to an association of three or more homologous chromosomes.

In *Pyrus malus* the number of bivalents is also higher in triploids than in diploids (NEBEL, DARLINGTON and MOFFETT, cited from DARLINGTON, 1937, p. 207). However, triploid *Pyrus malus* differs from the corresponding form of *Phleum pratense* by showing a rather high frequency of trivalents, and in addition there are also larger associations.

The cytological conditions in *Phleum* are paralleled to a large extent by similar phenomena in *Solanum nigrum* (JØRGENSEN, 1928). This is a polyploid species, the chromosome number being $2n = 72$. Though irregularities were not entirely absent, meiosis in the diploid *nigrum* was generally characterized by an almost mechanical regularity. The first and second metaphase plates were always found to contain 36 chromosomes. At diakinesis there were probably 36 gemini, though the presence of a few trivalents or quadrivalents was not quite excluded. In haploid *nigrum* 36 univalents might be expected at meiosis, but the homology between the genomes constituting the species revealed itself by the formation of $12_{II} + 12_{I}$ in typical cases. No quite clear associations of three chromosomes were observed in the p. m. c., but in the megaspore mother cells a few trivalents could be distinguished. This indicates that *S. nigrum* contains three more or less homologous genomes (JØRGENSEN, l. c. p. 195).

The meiotic conditions in triploid *Solanum nigrum* are of still more interest with regard to our material. According to JØRGENSEN (l. c. pp. 181—183), most of the 108 chromosomes form bivalents, the number of elements present at I—M varying between 50 and 65. Since only bivalents and univalents seemed to occur, it was concluded that the three sets of 36 chromosomes combined in such a way that two of them formed 36_{II}, the chromosomes of the third set pairing inter se. Thus, the triploid *nigrum* behaved as if composed of a diploid and a haploid, the scheme $36_{II} + 12_{II} + 12_{I}$ being approached.

Evidently, the mode of meiosis in this triploid is of exactly the same type as in *Phleum pratense*. In both species trivalents are absent or at least rather rare. This is very striking, since the perfect homology

between the three sets of chromosomes would be expected to permit the union of all the chromosomes into trivalents. Thus, in these triploids other strong factors besides chromosome homology are of prime importance for the mode of chromosome association.

In other respects there are some differences between the triploids of the two species. The *Phleum* triploid is perfectly fertile, and in the progeny the chromosome number is never lower than 56. This is due to a general regularity of meiosis and a regular formation of tetrads. The triploid *Solanum nigrum*, on the contrary, was rather sterile, and at the tetrad stage from two to eight cells were formed. JØRGENSEN (l. c. p. 183) also mentions that according to WINKLER's breeding experiments (1922) the offspring from the triploids in a few generations turn into diploids.

Since the triploids in *Phleum pratense* and *Solanum nigrum* are both characterized by rather high chromosome numbers, it is of interest to compare these cases with other triploids having high chromosome numbers. Such material is, for instance, represented by triploid *Nicotiana Tabacum*, having $2n = 72$. Triploids of this kind have been examined by GOODSPEED (1930) and EAST (1933). GOODSPEED observed trivalents as well as bivalents and univalents at I—M, but the exact number could not be determined. Judging from the total number of elements at I—M, which was frequently higher than 36, the average number of trivalents was probably lower than 12. In his triploid EAST (l. c.) found that the number of chromosomal bodies at I—M varied from 36 to 42, the former number being most typical. Though a few of these bodies may correspond to univalents or trivalents, most of them are considered to be bivalents. If this is true, meiosis in triploid *Nicotiana Tabacum* would be rather similar to meiosis in triploid *Phleum pratense*. At any rate, the frequency of trivalents seems to be relatively low and far lower than the possible maximum of 24. — Meiosis has also been studied in autotriploid twin plants of *Triticum vulgare*, having $2n = 63$ just as our *Phleum* twins (YAMAMOTO, 1936). In this material, in contrast to *Phleum*, *Solanum* and *Nicotiana*, the frequency of trivalents seems to be rather high, but no exact counts of the number of trivalents were made.

In comparison with our observations in *Phleum* the cytogenetic results obtained by SHIMOTOMAI (1931, 1932, 1933) in hybridization experiments in *Chrysanthemum* are quite interesting. The meiotic behaviour in the *Chrysanthemum* hybrids studied seems to depend entirely on the chromosome number of the parents. If the difference in gametic

chromosome number between the species crossed is an even multiple
of the basic chromosome number 9 (generally 18), the resulting hybrid
will have perfectly regular meiosis with bivalents only. If the numerical
difference between the parents is an uneven multiple (generally 9), the
supernumerary chromosomes cannot find any mates and as a rule
appear as univalents at meiosis. A cross 18×36 will consequently
give a hybrid with $2n = 54$, which chromosomes form 27 bivalents at
meiosis. A cross of the type 18×27, on the contrary, gives an F_1 with
$18_{II} + 9_I$ at meiosis. In hybrids of the first kind all gametes receive
the same chromosome number, and such hybrids therefore behave as
new cytologically constant species in contrast to hybrids of the second
kind. In the *Chrysanthemum* material studied the different genomes,
judging from the mode of pairing, must be largely homologous and may
be recombined at will without serious disturbances. The behaviour of
the hybrids seem to depend on purely numerical conditions. These
results are quite similar to those found e. g. in *Papaver* (LJUNGDAHL,
1924) and in *Fragaria* (LILIENFELD, 1933). For other similar cases cf.
DARLINGTON, 1937, p. 208.

The absence of multivalents in the pure species or balanced hybrids
Thus, though the genomes of the polyploid *Chrysanthemum* species
in question are largely homologous, there are no multivalents at meiosis
in the pure species or in balanced hybrids derived from them. In
hybrids with univalents, however, trivalents may also occur. In the
Chrysanthemum hybrid *lavandulaefolium* $\times$ *indicum* the frequency of
trivalents was even found to be rather high (TAKEMOTO, 1939), con-
figurations such as $8_{III} + 1_{II} + 1_I$ and $5_{III} + 4_{II} + 4_I$ being observed. Of
the two parent species, *C. lavandulaefolium* is diploid $(2n = 18)$ and
C. indicum tetraploid $(2n = 36)$. Though the two genomes of *indicum*
must be largely homologous, which was also verified by chromosome
morphological studies (SHIMOTOMAI and TAKEMOTO, 1939), they do not
seem to form quadrivalents in pure *C. indicum* (cf. SHIMOTOMAI, 1933,
Fig. 1 g).

The absence of multivalents in the pure species or balanced hybrids
seemed peculiar at a previous discussion of the *Chrysanthemum* results
(MÜNTZING, 1936), but in view of the present results in *Phleum* it is
more easy to understand. It is not necessary to assume a condition of
differential affinity, the absence of multivalents may simply be the
result of a strong tendency to two-by-two pairing of the same kind as in
Phleum. If this tendency is at work, the »need» of association is satis-
fied by the pairing of two homologous chromosomes. Even if there are

groups of four perfectly homologous chromosomes, these will pair as two bivalents and not as one quadrivalent.

In *Solanum nigrum* not only haploids and triploids were studied by JØRGENSEN (1928) but also tetraploids. Considering the remarkably regular I—M association in the triploid, it is not surprising that in the tetraploid (2n = 144) multiple associations were quite rare, the great majority of the chromosomes forming bivalents. Only a few quadrivalents were observed at diakinesis and may also occur at first metaphase.

In *Phleum pratense* tetraploid forms (2n = 84) have not yet been produced, but their production is probably only a question of time. They might either be produced by colchicine treatment of ordinary timothy or by selection of twin seedlings in the progeny of 8x plants. As described in detail in this paper, many plants having exactly or approximately the 8x chromosome number 56 were obtained in the offspring of the triploid twins. By raising new twin seedlings from 56-chromosome plants it should be relatively easy to get plants with 56 + 28 = 84 chromosomes. These would be tetraploid in relation to ordinary *Phleum pratense* (2n = 42). By crosses between plants with 84 and 56 chromosomes it should also be possible to raise individuals with 2n = 70. In view of the strong tendency to two-by-two pairing in *Phleum*, it may be predicted that such plants should in the main have a regular meiosis with 35 bivalents. In the same way the tetraploid individuals (n = 84) should mainly have 42 bivalents at diakinesis and first metaphase. — Starting again from plants with 70 and 84 chromosomes, it should be possible to produce true breeding strains with still higher chromosome multiples.

Pending the production of such new strains, the mode of chromosome association in ordinary *Phleum pratense* may be considered once more from another point of view. As observed by MÜNTZING (1935) and as described in the present paper, the typical I—M configuration is 21_{II}. The absence or rarity of multivalents would seem to indicate that the three genomes of *Phleum pratense* are well differentiated. However, the results of NORDENSKIÖLD (1937) and ourselves strongly indicate that at least two of the three genomes (A_1 and A_2) are highly homologous. This was evident, firstly by the regular formation of 14 bivalents in the hybrid between *Phleum pratense* and *nodosum* and, secondly, by the formation of 28 bivalents (+ 7 univalents) in our triploid *pratense.* It should also be observed that the tetraploid *pratense* × *nodosum* hybrid (2n = 28) as well as triploid *pratense* (2n = 63) have

very good fertility, and that in the offspring of triploid *pratense* variation in plant vigour was not greater than in ordinary *Phleum pratense*.

The absence of quadrivalents in *P. pratense* does not necessarily imply that the bivalents formed are always of the type A_1—A_1 and A_2—A_2. It may be that pairing is just as frequent between the A_1 and A_2 chromosomes. Among the four homologous genomes present the pairing tendency is completely satisfied by the formation of bivalents. It is quite possible that these bivalents are formed at random, and if that is true, the distinction between A_1 and A_2 is no longer valid. Thus, it seems possible and even probable that the genome formula of *P. pratense* should be given as *NN AA AA* instead of *NN A_1A_1 A_2A_2*.

Studies of segregation ratios in *Phleum pratense* would be important in order to decide if the above hypothesis of random pairing is correct. If there is no distinction between A_1 and A_2 genomes, dihybrid ratios for factors located in these genomes would be of the autotetraploid type 35 : 1 rather than 15 : 1. Unfortunately, as far as we know, distinct segregation ratios have not yet been described for this species.

Such ratios, however, are known in *Solanum tuberosum*, in which the cytogenetic conditions seem to be similar to those in *Phleum pratense*. LUNDEN (1937) studying the genetics of the species found autotetraploid segregation ratios for several factors. This is remarkable since *S. tuberosum* has generally been assumed to be an allotetraploid species. All European potato varieties have 2n = 48, and at meiosis the great majority of these chromosomes form bivalents, only a small number forming true multiple associations (BLEJER, 1931; MEURMAN and RANCKEN, 1932; ELLISON, 1936). Under such circumstances it must be concluded that in the potato the four genomes of 12 chromosomes are highly homologous, and that there is a random two-by-two pairing between the homologous chromosomes of all four genomes. *Solanum tuberosum* like *Phleum pratense* may, indeed, be one of those species in which four homologous chromosomes prefer to associate at random as two pairs instead of forming a quadrivalent.

This possibility is further strengthened by the cytological observations of LAMM (1938) and PROPACH (1937, 1938). LAMM studied a polyhaploid potato plant (2n = 24), being a member of a pair of twin seedlings in the progeny of a tetraploid F_1 hybrid (2n = 48) between triploid *Solanum chaucha* (from South America, 2n = 36) and tetraploid *S. tuberosum* (2n = 48). In this haploid the 24 chromosomes were associated at I—M as 12 bivalents in the majority of the p. m. c., and many of these bivalents were joined by two chiasmata. In the

tetraploid sister twin most of the chromosomes as usual formed bivalents, though some trivalents and quadrivalents were also present. The average chiasma frequency per chromosome was found to be 1,22 in the haploid, 1,27 in the tetraploid.

Thus, it seems clear that *Solanum tuberosum* belongs to the same group of material as *Solanum nigrum, Phleum pratense,* some *Chrysanthemum* and *Papaver* species, etc., for which a high degree of autopolyploidy is characteristic though this is not apparent at meiosis in the pure species.

The number of *Solanum* species showing such conditions are not limited to *S. nigrum* and *tuberosum*. PROPACH (l. c.) has studied some other polyploid *Solanum* species, showing quite regular bivalent formation in the pure species but autosyndesis in hybrids with diploid species. PROPACH (1937) concludes that the supposed allopolyploidy of *S. acaule* and *demissum* is only apparent, their genomes in reality being homologous. Though autopolyploid the species are supposed to have acquired, during their phylogeny, the capacity of regular bivalent formation.

According to our opinion, and in agreement with PROPACH's conclusions, *the results in Phleum and the other genera, showing similar conditions, can only be explained by assuming a special genotypically controlled tendency to bivalent formation. In species capable of such a control the formation of multivalents is prevented, and thus a regular meiosis is secured in spite of complete or almost complete homology between more than two genomes.* — By assuming such a force we do not deny the prime importance of differential affinity and chiasma frequency for the mode of chromosome pairing (cf. DARLINGTON, 1937), but these factors do not seem to be sufficient to explain the whole story. — In presumably autotetraploid *Tulipa* species UPCOTT (1939 b) finds a low frequency of quadrivalents, and this is correlated with a lower chiasma frequency and fewer changes of partner at pachytene than in diploid and triploid tulips. Judging from these observations, the specific genes preventing multivalent formation may excersize their influence either by a reduction in the frequency of chiasmata in the regions paired or by a reduction of the changes of partner at pachytene. The latter method may be the more important one for the cases discussed in this paper. In *Phleum*, at least, the chiasma frequency was high enough to permit the formation of multivalents.

From the facts described above it is clear that some species, though having a regular meiosis without multivalents, are nevertheless highly

autopolyploid. Thus, though *the presence of multivalents at meiosis may indicate autopolyploidy, the absence of multivalents does not prove the species to be allopolyploid.* In a paper on the evolutionary significance of autopolyploidy one of us (MÜNTZING, 1936) presented evidence strongly indicating that the rôle of autopolyploidy in nature had been underestimated. This was especially evident from an analysis of the properties of a series of intraspecific chromosome races and closely related species representing different degrees of polyploidy.

In most of the cases studied the polyploid forms were characterized by the presence of multivalents at meiosis, but in a minority of cases there were no multivalents in the polyploid forms. In some of the latter cases (*Nasturtium, Chrysanthemum* and *Betula*) other evidence than the presence of multivalents strongly indicated autopolyploidy, and therefore the conclusion was drawn (l. c. p. 313) that »though a few cases occur, in which the absence of multivalents indicates allopolyploidy, these are only exceptions and in some cases even uncertain exceptions to the rule that polyploid intraspecific chromosome races are generally autopolyploid».

This conclusion is further supported by the evidence discussed in the present paper, firstly, because it is now quite clear that genomatic homology does not always lead to multivalent formation, secondly, because two of the apparent exceptions to the rule were represented by *Phleum alpinum* and *Phleum pratense-nodosum*. Considering the *Phleum* results presented in this paper and those obtained by NORDEN-SKIÖLD (1937), it is quite evident that the *Phleum* species in question contain genomes that are completely or partially homologous. Thus, the exceptions to the rule that polyploid intraspecific chromosome races are autopolyploid now seem to be still fewer than in 1936.

It is also evident that the occurrence of autopolyploidy is not limited to polyploid chromosome races but is also met with in polyploid species. So far only such species as are characterized by multivalents at meiosis have been recognized with certainty as being autopolyploid (cf. MÜNTZING, 1936, p. 334). However, the examples in the genus *Solanum*, for instance, demonstrate that this category also includes species with only bivalents at meiosis. Thus, *races and species, which are completely or predominantly autopolyploid, must be even more frequent than there was reason to assume a few years ago* [1].

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[1] In her recent paper on polyploidy in *Tulipa*, UPCOTT (1939 b) finds it appropriate to critisize the paper of MÜNTZING (1936) by confronting two detached and apparently contradictory sentences. However, anyone reading the whole chapter will

Though, from a theoretical point of view, the mode of chromosome pairing is of most interest in our triploid *Phleum* twins, some other points may also be briefly considered. — It is rather striking that in the progeny of the twins with $2n = 63$ the plants are but very slightly sensitive to the variation in chromosome number, plants with 59—61 chromosomes having about the same vigour as plants with 56—58 and 62—64 chromosomes. This independence of chromosome number is certainly due to the autopolyploid constitution and the rather high absolute chromosome number of the material. Quite analogous results were obtained by MÜNTZING (1937 b, 1940) in material of *Dactylis* and *Poa*. Also in allopolyploid *Triticale* strains deviations from the typical chromosome number 56 have not much effect on plant vigour (MÜNTZING, 1939).

The high degree of fertility in the *Phleum* material under discussion may be explained in the same way. All genes necessary for the functioning of the pollen grains and ovules are reduplicated, and therefore it does not matter whether there are any extra chromosomes present or not.

From a practical point of view this undisturbed fertility is of course very favourable, and the same is true of the regular meiosis in the triploid twins. Since there is every reason to believe that the same regularity will be repeated in the offspring, it should be an easy task to raise an unlimited number of stable strains, having $2n = 56$. Whether the other possible derivatives, having $2n = 70$ and $2n = 84$ etc., will be quite stable is so far an open question. — For breeding purposes it

not misunderstand the meaning. Since probably nothing can be said against the first statement, that the presence of multivalents indicates autopolyploidy, the absence of multivalents allopolyploidy, the other sentence critized may be considered, viz. »Thus, though a few cases occur, in which the absence of multivalents indicates allopolyploidy, these are only exceptions and in some cases even uncertain exceptions to the rule that polyploid intraspecific chromosome races are generally autopolyploid »

Since the evidence on which this statement is based is still perfectly valid, there is no reason for a change of opinion. On the contrary, the data discussed above in the present paper give further support to the view that polyploid intraspecific chromosome races are really in most cases autopolyploid.

In the paper by UPCOTT (l. c. p. 335) it is further stated, strangely enough in contradistinction to some quotations from the paper by MÜNTZING (1936), that many polyploid species are intermediate between the two extremes (presumably auto- and allopolyploidy). We perfectly agree with this opinion, especially since chapter VII in MÜNTZING's paper (l. c. pp. 361—365) deals with the same subject and reaches the same conclusions.

would be òf most interest to concentrate on the production of strains with 56 chromosomes. This program is further supported by the observation of a very slight, but nevertheless significant, correlation between chromosome number and vigour, the plants having the exact or approximate chromosome number 56 being somewhat superior to individuals with chromosome numbers intermediate between 56 and 63.

In spite of the presence of individuals of the latter kind some of the twin progenies studied gave a yield which was quite or almost as good as the yield of the standard (2n = 42). This is remarkable, since the standard used was a well-known commercial variety and the first triploid twins available were of unknown origin. Good future results may be expected a) by the production of 63-chromosome plants from the best *P. pratense* material available, b) by selection and intercrossing among the triploid twins thus produced, and c) by selection of strains with 56 chromosomes in the following generations. — Due to meiotic elimination the chromosome number in the offspring of 63-chromosome twins will in time probably automatically reach the 8x condition, but by a direct selection of vigorous plants with exactly or approximately 56 chromosomes the process may be greatly accelerated.

Finally, it may be mentioned that the change in chromosome number from 42 to 56—64 does not involve any obvious change in the chemical properties. Thanks to Dr. J. LINDBERG, Svalöf, determinations of water content, crude protein, crude fat, soluble carbohydrates, crude fibre and ashes were undertaken in the twin pairs, twin progenies and the standard variety. In all of these properties the material with high chromosome numbers had sometimes slightly higher, sometimes slightly lower values than the corresponding types with the normal chromosome number. Thus, the breeding of timothy with 56 chromosomes instead of 42 may evidently be undertaken without the risk of a decreasing chemical quality.

SUMMARY.

1. Twin plants of *Phleum pratense*, having 2n = 63 instead of the normal number 2n = 42, were found to have good vigour and perfectly good fertility. The ›triploid› members of the twin pairs have longer, broader and thicker leaves than the corresponding diploids and also thicker stems, longer and thicker culms, bigger spikelets and larger pollen grains.

2. Meiosis was studied in the p. m. c. In the triploids the fre-

quency of bivalents was much higher and the frequency of trivalents much lower than expected. The sum of the bivalents and the occasional trivalents was hardly ever lower than 28, and the number of univalents not higher than seven.

3. Estimating the degree of meiotic elimination the average chromosome number of the gametes formed by the triploids was found to be 30,03. All gametes will carry at least 28 chromosomes. In the offspring of the triploid twins chromosome counts were undertaken in 186 individuals. The chromosome number was found to range from 56 to 64, the average value being 59,66. Thus, the agreement between expectation and observation is quite good.

4. Plant weight was compared in the twin progenies and a standard variety having the normal chromosome number 42. Considering the chromosomal variation in the twin progenies, their average productivity was surprisingly good and about equal to that of the standard. In the first summer the twin progenies gave a higher yield than the standard, but in the later weighings the results were less favourable. Thus, the twin progenies were less hardy than the standard.

5. In the twin progenies the possible occurrence of a correlation between chromosome number and vigour was tested. Combining all data available, the plants having the exact or approximate 8x constitution were found to be slightly more vigorous than those having numbers intermediate between 56 and 63. Pollen fertility, on the other hand, was found to be quite good in plants with any chromosome number.

6. The mode of chromosome pairing in diploid and triploid *Phleum pratense* is discussed. Two of the three genomes of the species must be homologous, and thus the genome formula should be given as $NN\ A_1A_1\ A_2A_2$ rather than $NN\ AA\ BB$. It is even possible that A_1 and A_2 are identical, and that pairing between the A_1 and A_2 chromosomes is just as frequent as the pairing of the type A_1—A_1 and A_2—A_2.

7. In *Phleum pratense* the »need» of association is almost completely satisfied by the pairing of two homologous chromosomes, even if plenty of other homologous chromosomes are present in the nucleus. This is not due to an insufficient chiasma frequency but must be caused by a special, genotypically controlled tendency to bivalent formation. Similar cases in other genera are discussed.

8. Since an autopolyploid constitution is not always accompanied by multivalent formation, races and species which are completely or

mainly autopolyploid must be more frequent than there was earlier reason to assume.

9. The mode of production and practical importance of timothy strains with 56, 70 and 84 chromosomes are discussed.

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